A HISTO PATHOLOGICAL ANALYSIS OF PROSTATIC LESIONS AND ASSESSMENT OF PROGNOSIS WITH IMMUNOHISTOCHEMISTRY.

DISSERTATION

SUBMITTED FOR M.D. BRANCH III

(PATHOLOGY)

MARCH – 2009

THE TAMILNADU Dr. M.G.R. MEDICAL UNIVERSITY

CHENNAI - TAMILNADU

CERTIFICATE

This is to certify that this dissertation entitled **"A HISTOPATHOLOGICAL ANALYSIS OF PROSTATIC LESIONS AND THE ASSESSMENT OF PROGNOSIS WITH IMMUNOHISTOCHEMISTRY"** is the bonafide record work done by **DR. K. RAJESWARI** submitted as partial fulfillment for the requirements of **M.D Degree Examinations, Pathology** to be held in March 2009.

DR T.B.UMADEVI,M.D.,
Professor & Head of the Department,
Department of Pathology,
Thanjavur Medical College,
Thanjavur.

THE DEAN,
Thanjavur Medical College,
Thanjavur.

ACKNOWLEDGEMENT

I wish to express my sincere and profound gratitude to my **PROFESSOR DR T B UMADEVI** MD(Pathology) Professor and Head of the Department of Pathology,Thanjavur Medical College,Thanjavur,for her valuable guidance at every stage,constant encouragement and words of advice which have been the motivating forces in bringing forth this piece of work.

I am also extremely grateful to **Dr N ARUMUGAM** MD.,Reader of pathology who has offered many valuable suggestions and encouragement during this period,whose help has been invaluable to me.

My heartfelt thanks to **Dr A L SHANTHI** M D.,Assistant Professor for her valuable guidance and encouragement throughout my study.

I am also thankful to all my Assistant Professors for their valuable suggestions and guidance at every stage in this study..

I would also like to express my sincere thanks to my fellow-postgraduates,Lab technicians and all the staffs of the department for their generous help throughout my study.

Above all,I thank our **DEAN**,for granting me the permission to carry out this work.

CONTENTS

INTRODUCTION

DISEASES OF THE PROSTATE ACCOUNT FOR CONSIDERABLE MORBIDITY AND MORTALITY AMONG AGING POPULATION WORLDWIDE. Patients presenting with prostatic symptoms account for 22% of the total urology cases,and every 7 th patient will be presenting with urologic emergency.[12,59]

PROSTATITIS, BENIGN NODULAR HYPERPLASIA AND PROSTATIC ADENOCARCINOMA ARE COMMON DISEASES AFFECTING MEN COMMONLY AFTER THEIR FIFTH DECADE.[8,28,49]

There has been steadily increasing interest in prostatic pathology as reflected by numerous publications in the last decade.[28]

The clinical and basic scientific studies have led to a better understanding of previously recognized tumours and morphologic studies have described many new entities.[28,49]

The histopathologic spectrum of prostatic hyperplasia has been broadened with descriptions of several variants and work done primarily in the last two decades has considerably expanded our knowledge of the pathology and **BIOLOGY OF PROSTATIC ADENOCARCINOMA,IN PARTICULAR ITS POSSIBLE PRECURSOR LESIONS,SPECIAL HISTOLOGIC VARIANTS AND PROGNOSTIC FACTORS.**[25,37,44]

The **CLINICAL INCIDENCE** of nodular hyperplasia is only 8% during 4 th decade,but it **REACHES 50% IN THE 5 TH DECADE AND 75% IN THE 8 TH DECADE**.It has been estimated that the process begins before the age of 30 and that its doubling time progressively increases from the early to late stages.[37,44,53,55]

PROSTATIC CARCINOMA IS THE LEADING CAUSE OF NEWLY DIAGNOSED CANCER IN WESTERN MEN and second only to lung cancer as a leading cause of cancer related deaths in men.[28]

THE AGE ADJUSTED INCIDENCE IS ON THE RISE IN MOST COUNTRIES INCLUDING INDIA.[28,55,41]

Almost **75%** of the men diagnosed with **PROSTATIC CARCINOMA** are aged **65 OR OLDER**,but the tumours can be seen even in children and adolescents though,very rarely.[11,12,21,28]

Normal and benign glands may contain neutral mucosubstances whereas invasive carcinomas and precursor lesions may harbour acid mucins.Thus mucin histochemistry may be used as an ancillary method for diagnosing diseases of prostate.[16,20,24,36]

THE NUMBER OF MITOTIC FIGURES INCREASED PROGRESSIVELY FROM BENIGN EPITHELIUM THROUGH PRECURSOR LESION TO MALIGNANCY.HENCE,ELEVATION IN THE PROLIFERATION INDICES OF KI – 67 APPEARS TO REFLECT PROGRESSION AND IS BECOMING AN IMPORTANT DIAGNOSTIC AND PROGNOSTIC FACTOR.[4,12,13,32,33,45,49,53,55,59]

This study is undertaken in view of evaluating the actual incidence of different prostatic lesions in semi urban area like Thanjavur with particular attention to proliferative status of precancerous and different grades of frankly malignant lesions.In addition, the recent literatures,journals and research publications regarding prostatic pathology are also immensely reviewed.

AIM OF THE STUDY

1) To analyse the incidence of various prostatic lesions in our institution during 2006-2008.
2) To sub classify the lesions and their prevalence.
3) To **SUBTYPE THE NEOPLASTIC LESIONS**.
4) To exactly **DIFFERENTIATE THE PROSTATIC CARCINOMA MIMICKERS FROM WELL DIFFERENTIATED CARCINOMAS**.
5) To **APPLY UPGRADED GLEASON'S TERTIARY GRADING SYSTEM FOR PROSTATIC CARCINOMA**.
6) To study **THE NATURE OF MUCIN EXPRESSION IN VARIOUS PROSTATIC LESIONS.**
7) To study and **COMPARE THE PROLIFERATIVE ACTIVITY WITH IMMUNOHISTOCHEMICAL MARKER KI-67 IN BENIGN, PRECURSOR AND INVASIVE LESIONS OF PROSTATE.**

MATERIALS AND METHODS

This prospective study includes 120 cases of prostatic lesions,referred from **UROLOGY DEPARTMENT OF THANJAVUR MEDICAL COLLEGE,THANJAVUR,DURING THE TWO YEAR PERIOD FROM JUNE 2006 TO MAY 2008.**

A detailed clinical history like age,duration of complaints,nature of symptoms and haematological investigations were done in all cases.

All specimen we received were **TURP** specimen.Every tissue was carefully examined for the presence of yellow and/or hard areas and necrotic areas.As per the literature if all fragments received in a single container,all of the specimen submitted until four cassettes filled and if in excess,one additional cassette for each additional 10 gram of tissue.

In our cases,all specimen ranged in volume from 1 to 10 grams.Fixation was done by neutral buffered 10% formalin and the fixation period was 12 hrs.After adequate fixation,the entire specimen was submitted for processing.

Tissue processing was done with automated tissue processor and sections were made manually with histokinette of thickness 2 to 4 microns.Staining was done with routine haematoxylin and eosin and every slide was examined thoroughly and looked for the presence of malignancy,prostatic Intraepithelial Neoplasms,Atypical Adenomatous Hyperplasia,metaplasia,acute and chronic inflammation and other secondary changes associated with benign nodular hyperplasia.

To study the mucin histochemistry in benign,precursor and invasive lesions,Alcian blue at pH 2.5 and **PAS** were used.**ALCIAN BLUE** was the best available routine method to demonstrate acidic,weakly sulphated or acid-non sulphated mucosubstances when used at pH 2.5.

With PAS, it was accepted that acidic mucosubstances usually give a negative result.

IMMUNOHISTOCHEMISTRY WITH PROLIFERATIVE MARKER KI 67 was done for 10 cases which included Nodular Hyperplasia,prostatic intraepithelial neoplasia,low and high grade carcinomas.Randomly 100 cells per samples were examined and the number of positive cells with nuclear staining were noted**. KI – 67 LABELLING INDEX (KI – 67 LI)** expressed as percentage of KI – 67 positive cells.

REVIEW OF LITERATURE

Diseases of the prostate gland are common afflictions of men. In particular,prostatitis, Benign Nodular hyperplasia and prostate cancers are remarkably prevalent clinical disorders [32,48,53,55]

The human prostate gland is one of the male accessory sex organ which include the prostate, seminal vesicle and bulbourethral glands [32, 49, 47]

ANATOMY

The prostate gland is situated in the true pelvis between the bladder neck and urogenital diaphragm. It lies behind the inferior aspect of the symphysis pubis and anterior to the rectum. The prostate gland in young adults weighs about 20g and measures 4X3X2 cms. While the weight of the prostate gland has traditionally been thought to be relatively constant between the ages of 20 and 50, more recent autopsy date suggest a stepwise increase in prostatic weight with each decade of life.[49,53]

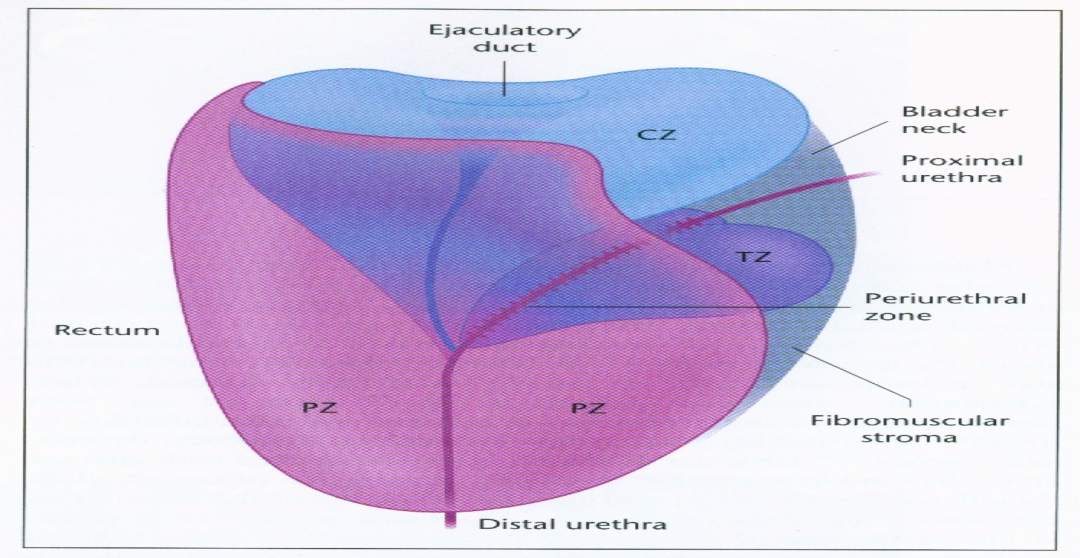
The prostate gland has an inverted conical shape with the base, the region of attachment of the seminal vesicles, in direct continuity with the bladder neck and the apex with the urogenital diaphragm. The prostate is organized around the first part of the urethra. Two ejaculatory ducts traverse the prostate. [32, 41, 47, 49]

ANATOMIC MODELS

Since, the early part of this century several models have been proposed. The one proposed by Mc Neal have now largely replaced previous models. In Mc Neal model, the key reference point is the prostatic urethra which has a 35 degree angulation in the midportion creating proximal and distal segments of about equal length. The verumontanum is part of the point of the distal urethral segment and consists of a bulge at the point of angulation.Glandular zones are arranged in relation to the prostatic urethra and ejaculatory ducts [25, 41, 49]

Mc Neal's model divides the prostate in to the three zones, the transition zone, peripheral zone and central zone each of which has a different glandular organization and proclivity for disease [25, 41, 49, 59]

FIGURE 1



- **Transition zone - 5 % of volume, chief source of NH, 15-20% of carcinomas ,**
- **Central zone - 25% of volume, 10% carcinomas ,**
- **Peripheral zone -70% of Volume, 70-75% of Carcinomas[49]**

HISTOLOGY

The prostate gland has a compound tubuloalveolar organization with some variation in histology in the three zones. The acinar units responsible for prostatic secretion open in to a series of ramifying ducts which empty in to large primary ducts, which open in to the prostatic urethra.[9,11,49]

The prostatic acini and ducts, except for the immediate periurethral segment, are lined by a double-layered epithelium consisting of secretory and basal cells. The secretory cells are cuboidal to columnar. They generally have basally located, uniform nuclei and abundant apical cytoplasm which is pale and granular. These cells are responsible for the production and release of biochemical products.The secretory cells rest on a basal cell layer which consists of cuboidal to flattened attenuated cells with relatively high nuclear to cytoplasmic ratios. The basal cells rest on a delicate basement membrane which is not usually visualized with routine stains. A population of stem cells resides within the basal cell compartment. [41, 49, 59]

Prostatic secretion is commonly present at least focally, which is PAS positive. Neutral mucin may also be seen in prostatic adenocarcinoma. Acidic mucins are not usually found in normal or hyperplastic glands but are commonly found, at least focally, in prostatic adenocarcinoma [16, 49]

Corpora amylacea are laminated luminal concretions that are commonly present in prostatic glands and increase in prevalence in prostate with advancing age [25]

The prostatic ducts and acini are surrounded by stroma consisting of smooth muscle cells, fibroblasts and undifferentiated spindle cells embedded in a matrix consisting of collagen and ground substance.In addition, prostate gland also has neuroendocrine cells, lipofuscin and melanin pigments. [11, 32, 48]

With advancing age, the prostate gland is commonly involved by both atrophy and hyperplasia.

HISTOCHEMICAL FEATURES

Normal and hyperplastic prostate glands may contain neutral mucosubstances, both neutral and acid mucopolysaccharides are found predominantly in neoplastic disease including both prostatic intraepithelial neoplasia and adenocarcinoma. Mucin studies will confirm the mucinous nature of the often blue-tinged secretions frequently seen in neoplastic acini.Usually only small amount of mucin is identified within the lumens.[19, 20, 23, 36]

Elbadawi et al reported that neutral mucins may be secreted in small amount by prostatic epithelial cells whether benign or malignant and they are present in cytoplasmic granules. They are variably eosinophilic and are stained by the PAS technique.[16, 20, 23, 36, 51]

Frank et al and hukill et al in their independent studies stated that acid mucins are rare and scant in benign prostate gland lumina and their abundant secretion is specific for mucinous areas of prostatic cancer. They are usually faintly basophilic with H&E stain, but they are specifically identified by Alcian blue technique which does not stain the neutral mucin .[20,23,36]

Morphological and histochemical analysis by Mc Neal revealed mucin secreting areas in 33% of prostatic adenocarcinoma.Taylor reported that the presence of acid mucin is of diagnostic significance.Ro et al agrees that the stains for acid mucopolysaccharides are useful in confirming the diagnosis of prostate cancers especially in equivocal cases .[16,20, 23].

IMMUNOHISTOCHEMICAL FEATURES

The secretory cells and the basal cells stain with a variety of keratin antibodies. The basal cells in addition, demonstrate positivity with antibodies against high molecular weight keratin.The antibody designated 34BE12 has specificity for high molecular weight cytokeratins and has been widely used to demonstrate basal cells, which is important diagnostically. The complete absence of basal cells in an acinar proliferation is an important criteria for malignancy and is useful in separating small acinar paterrns of carcinoma from carcinoma mimickers such as atrophy, postatrophic hyperplasia, radiation induced atypia and atypical adenomatous hyperplasia.[11, 13, 19, 25,28]

PROSTATE SPECIFIC ANTIGEN (PSA) & PROSTATIC ACID PHOSPHATASE (PAP)

Glandular epithelia from all prostatic zones stain with PSA.. Most adenocarcinomas display positivity with PSA antibodies, although there is considerable irtertumoural and intratumoural variation in extent and intensity .[11, 13, 32, 41, 49].

PAP is found in normal and hyperplastic and also in most prostatic adenocarcinomas. The histochemical assessment of PAP is limited because of its non specificity. Antibodies to PAP and PSA are useful in adenocarcinomas of unknown origin and in poorly differentiated carcinomas of the lower genitourinary tract where the differential diagnosis includes prostatic adenocarcinoma and urothelial carcinoma [11, 13, 25, 49]

WHO CLASSIFICATION OF PROSTATIC HYPERPLASIA, TUMOURS AND TUMOUR LIKE LESIONS

Hyperplasia

- Benign nodular hyperplasia
- Basal cell hyperplasia
- Post atrophic hyperplasia
- Atypical adenomatous hyperplasia (adenosis)

Benign epithelial tumors

- Adenoma
- Multilocular cystadenoma

Prostatic intraepithelial neoplasia

Conventional adenocarcinoma

Special Variants of adenocarcinoma and other carcinomas

- Prostatic duct adenocarcinoma
- Mucinous (colloid) adenocarcinoma
- Signet ring cell carcinoma
- Adeno squamous carcinoma
- Squamous cell carcinoma
- Basaloid and adenoid cystic carcinoma

- Transitional cell carcinoma
- Small cell carcinoma
- Sarcomatoid carcinoma
- Lymphoepithelioma-like carcinoma
- Undifferntiated carcinoma, not otherwise specified.

Mixed Tumours

- Epithelial-stromal tumour ,not otherwise specified
- carcinosarcoma.

Mesenchymal tumours:

- Benign
- Leiomyorma
- Fibroma
- Hemangioma
- Others

Malignant

- Rhabdomyosarcoma
- Leiomyosarcoma
- Stromal sarcoma
- Others

Other tumours

- Hematolymphoid neoplasms
- Paraganglioma
- Germ cell tumours
- Willm's tumour

Secondary tumours

Tumour-like lesions.

BENIGN NODULAR HYPERPLASIA

Benign nodular hyperplasia represents nodular expansion of prostatic glandular elements, stromal elements, or both.[32, 49]The prevalence of nodular hyperplasia in austopsy series increases with advancing age from about 8% in the fourth decade to almost 90% at 80 years.In a clinical series of 1000 cases of prostatic hyperplasia, the youngest patient was 40 years age, and 98% were over 50 years.[47, 53,55]

The etiology and pathogenesis of nodular hyperplasia remain poorly understood. A number of factors, including marital status, socioeconomic status, libido, and diseases such as diabetes mellitus, hypertension, and cirrhosis, have been investigated and are not thought to be etiologically related to nodular hyperplasia. Testicular androgen production is necessary for the development of nodular hyperplasia. [47, 59]

The principal androgenic hormones are testosterone and dihydrotestosterone(DHT). DHT is the active metabolite of testosterone which many believe is related to the development of nodular hyperplasia [47, 59]

Nodular hyperplasia preferentially involves the proximal periurethral tissues (the so called estrogen sensitive zone). The transition zone expands with advancing age, resulting in a nodular gross appearance due to the lobar architecture of this zone; most clinically significant nodules develop here. [25, 32, 49]

It is likely that the pathogenesis of nodular hyperplasia is multifactorial and involves hormonal alterations,stromal-epithelial communication and fundamental changes in proliferative cellular compartments. [49,50]

CLINICAL FEATURES

About 50% of patients with grossly enlarged glands develop prostatism. The symptoms may be categorized as either obstructive or irritative. Obstructive symptoms include hesitancy, weak stream, and terminal dribbling. Irritative symptoms include pain,increased frequency and dysuria.

The severity of symptoms does not necessarily correlate with gland size. Patient may present with acute urinary retention resulting from complete blockage. This situation present a urologic emergency. [53,59, 65] On digital rectal examination, the prostatic gland in typically large and may have a nodular contour. The enlargement is often symmetric, and the texture is rubbery. Tenderness may be present, especially if there is superimposed prostatitis. [49]

FRANK'S CLASSIFICATION OF HYPERPLASTIC NODULES. [49]

- The stromal (fibrous or fibrovascular) nodule,
- The fibromuscular nodule,
- The muscular nodule,
- The fibroadenomatous nodule,
- The fibromyoadenomatous nodule.

The glands of nodular hyperplaisa are usually medium to large, sometimes cystic and may show architectural complexity and papillary infoldings. The epithelium usually has distinct double layer of secretory and basal cells, but the basal cells are not always conspicuous. The cells are often thrown in to papillary folds with some stratification, although the nuclei are usually aligned in a single row. [49, 53, 55]

SPECIAL HISTOLOGIC PATTERNS: [49]

1) Small glandular hyperplasia:.

Circumscribed proliferation of relaively small uniform glands in a fibramuscular stroma.

2) Cribriform hyperplasia:

Epithelial component consists of medium to large glands with a cribriform architecture. The cells may have abundant clear cytoplasm ("clear cell cribriform hyperplasia")

3) Leiomyomatous nodules:

Stromal nodules with prominent smooth muscle differentiation often cellular with mild nuclear variability.

4) Fibroadenoma like hyperplasia:

Glands and cellular fibrovascular stroma are organized in a fashion similar to fibroadenoma of breast.

5) Phyllodes-type hyperplasia:

In exuberant, sometimes myxoid, stromal proliferation, usually associated with intraluminal polypoid projections like phyllodes tumour of the breast.

SECONDARY CHANGES AND OTHER ASSOCIATED FINDINGS:

Sclerosing adenosis, cystic dilatations, corpora amylacea, psammomatous calcification, oncocytic or mucinous metaplasia, cystic dilatation, acute and chronic inflammation, basal cell hyperplasia, squamous metaplasia, transitional metaplasia, infarction are at times noted. PIN and atypical adenomatous hyperplasia may be seen in association with nodular hyperplasia. [37, 44, 53]

BASAL CELL HYPERPLASIA:

Typically found in the transition zone, but may also be atrophy associated and found in the peripheral zone. The nodule of nodular hyperplasia may be partially or completely involved. The basal cells are multi layered and relatively uniform, with a high nuclear to cytoplasmic ratio. When well delineated, the term basal cell adenoma has been used. [48, 53, 55]

Classification of prostatic basal cell hyperplasia: [49, 55, 47]

- Complete
- Incomplete
- Atypical basal cell hyperplasia
- Adenoid cystic-like basal cell hyperplasia
- Atrophy associated basal cell hyperplasia

The differential diagnosis are transitional cell metaplasia, prostatic intra epithelial neoplasia, conventional adenocarcinoma and basaloid carcinom[47, 49, 55]

PROSTATIC INTRAEPITHELIAL NEOPLASIA(PIN)

PIN is usually characterised by intraluminal proliferation of secretory epithelium that displays a spectrum of changes culminating in those that are indistinguishable from carcinoma. Occasionally, there is cytologic atypia without increased cellularity. High grade PIN(HGPIN) is strongly associated with invasive adenocarcinoma, particularly of the peripheral zone. [28, 45, 53]

There is much evidence supporting a preneoplastic role of PIN. The prevalence of PIN increases with age, peaking in the 6^{th} decade and predating the onset of most carcinomas by more than 5 years. PIN is much more common in prostates with carcinoma than in benign glands and is more often multifocal, more extensive and of higher grade in the former . Like carcinoma, PIN is mainly identified in the peripheral zone and is often adjacent to carcinoma. [28, 49, 59]

The extent of PIN is inversely related to the tumour stage, this may be related to replacement of areas of PIN by invasive tumour. As in carcinoma, increased production of acid mucin has been demonstrated.

The cytologic appearance of the cells of high grade PIN are similar to those seen in invasive carcinoma.Increased numbers of mitoses and apoptotic bodies, compared to benign glands have been identified, although in general there is a paucity of mitotic figures in PIN. [28, 49, 53,55]

In high grade PIN there is a partial loss of the basal cell layer while in carcinoma there is a complete loss. On occassion, carcinoma may be seen arising directly form an area of high grade PIN, a phenomenon that led MC Neal to propose the concept of microinvasion in association with high-grade PIN. [49,59]

There are no specific associated clinical features. Some patients present with elevated prostate-specific antigen levels, but this is likely often due to undetected invasive carcinoma. [59]

Mc Neal and Bostwick initially proposed a three grade system based on progressive abnormalities in architecture and cytology .Later, it was agreed to divide the process in to two grades, low and high. PIN 1 is now considered low grade (**LGPIN**) and PIN 2 and 3, high grade(**HGPIN**). [4, 10, 28, 49]

LOW GRADE PIN (LGPIN): Glands with crowding, stratification, irregular spacing of secretory cells. The nuclei are enlarged and there is anisonucleosis with usually inconspicuous nucleoli .[28,49]

HIGH GRADE PIN (HGPIN): Characterized by cellular proliferation within medium to large glands characterized additionally by cytologic atypia. Regardless of the architectural pattern, nuclear changes are the hall mark of high grade PIN. Nuclei are enlarged with hyperchromasia and irregular chromatin. Nucleoli are large, irregular,often multiple and may be focal(Grade 2PIN) or more extensive(Grade 3 PIN). [28,49,55]

HISTOLOGIC SUBTYPES OF PIN: [28,49]

1) Tufted
2) Micropapillary
3) Cribriform
4) Flat
5) Others:
    - Epithelial arches
    - Trabecular bars
    - Roman bridges
    - Solid patterns.

The basal cell-specific, high molecular weight keratin demonstrates a complete absence or discontinuous layer, a feature important in the differential diagnosis with prostatic carcinoma. [4,13]

CLINICAL SIGNIFICANCE OF PIN

The finding of low-grade PIN should not prompt further investigation and it is not reproducibly recognized. In contrast, high grade PIN is an important diagnosis, but should be made with care. If high-grade PIN is encountered in a needle biopsy specimen, additional levels should be considered to rule out carcinoma. A diagnosis of high-grade PIN without concurrent carcinoma should prompt careful clinical follow-up and further biopsies, especially if the serum PSA is elevated or abnormalities are noted on rectal or ultrasound examination. [7,10,18, 41]

ATYPICAL ADENOMATOUS HYPERPLASIA (ADENOSIS)

AAH is characterized by proliferating small to medium-sized acini that usually form a well-circumscribed nodule but occasionally extend in to the adjacent prostatic stroma. This has been identified in 1.5 to 19.6% of transurethral resectates and in up to 33% of radical prostatectomies. [8, 12, 32]

The evidence associating AAH and carcinoma is mainly circumstantial. The age of patient with AAH in usually 5-10 years less than that of carcinoma. AAH reportedly occurs more commonly in glands harboring cancer than in benign ones. Since AAH is often seen in transition zone, it has been suggested that it may be related to adenocarcinoma developing at this site. [28, 49, 55]

The basal cell-specific HMWK usually shows a discontinuous pattern. Basophilic mucin is rare, but when present is positive with the alcian blue stain. [4, 13]

From a therapeutic perspective, AAH is considered benign and patients should be followed conservatively. [12]

CARCINOMA OF THE PROSTATE

Establishing, or ruling out, the diagnosis of carcinoma of the prostate has been a well-known challenge for pathologists for many years. In the western countries, prostatic adenocarcinoma is the most common noncutaneous malignant neoplasm in humans, and is second only to lung cancer in mortality. Both incidence and mortality rate have increased over the last few decades all over the world, even in India. [11, 19, 28]

Clinically manifest prostatic cancer usually occurs in the sixth to eighth decades and is uncommon in patients under 50 years of age. There is considerable geographic variation in the incidence and mortality rates for prostatic cancer. Low rates are seen in the far east and very high rate in Northern European and North American populations. [19, 28, 49]

The etiology and pathogenesis of prostate cancer are poorly understood. Both genetic and epigenetic factors have been implicated. There is a well-documented familial association in a minority of cases. The strongest risk factors are advancing age, race, heredity and hormonal activity. The other doubtful factors are viruses, cadmium exposure, high fat diet, vitamin A and D deficiency etc. [41, 49, 55, 59]

CLINICAL FEATURES:

Most often asymptomatic, but locally extensive tumours cause pubic pain, rectal obstruction or bleeding. Presenting symptoms of metastatic disease include bone pain and tenderness, cord compression. Occasionally, enlarged lymphnodes may be the presenting symptoms. On rare occasions, prostatic carcinoma may manifest as a paraneoplastic syndrome. [8, 11, 12, 55]

Screerning methods for prostatic carcinoma are Digital rectal examination (DRE), measurement of serum PSA, transrectal ultra sound (TRUS). The detection rate for DRE is about 0.8-2.7%. But, most carcinomas detected by transurethral resection and in PSA screening programs are not palpable. [49, 55, 59]

In many patients, abnormal PSA elevation may be the first finding. A PSA value above 4 ng/ml is considered abnormal. 4-10 ng/ml -low range abnormality>10ng/ml- High range abnormality .A variety of PSA indices have been developed, including age-specific PSA tables, PSA density, PSA velocity. [8, 11, 59]

On TRUS, areas of hypoechogenicity may be detected. But in some cases no abnormalities are seen. Directed biopsies of abnormal areas can be obtained, and if no abnormalities are detected, multiple areas are systematically biopsied. Commonly a sextant approach is undertaken ,biopsies form the basal, mid and apical portions of the gland, with or without the transition zone, are taken on both sides. [12, 19, 21, 49]

PATHOLOGIC FEATURES:

The goal of the pathologic examination is to provide the necessary information to guide further therapy and assess the prognosis. Prostatic carcinomas can be divided in to two major categories:

1) Adenocarcinoma of peripheral ducts and acini.
2) Carcinoma of large ducts.

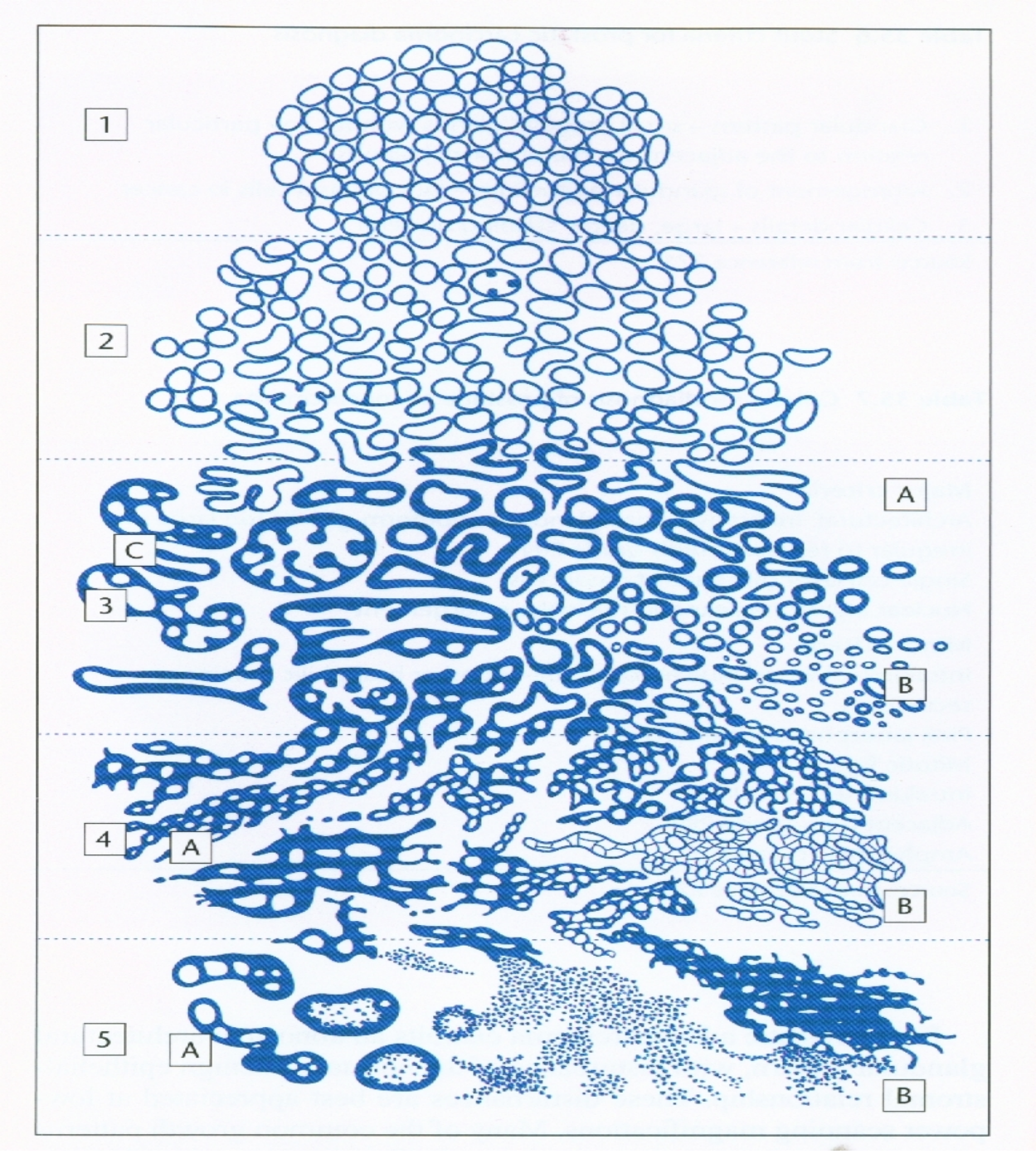
Recently, it is proposed that, it is the site of the growth, rather than the origin that governs the tumour architecture. [8, 11, 19]

Most prostatic carcinomas arise in the peripheral zone. Grossly the tumour may be difficult to see but usually can be identified as gray or yellowish, poorly delineated, firm area.Microscopically,prostatic adenocarcinomas exhibit a wide spectrum of appearances, ranging from anaplastic tumours to highly differentiated neoplasms which will be very difficult to differentiate from normal and benign glands. [11, 21, 32]

The accurate diagnosis of prostatic adenocarcinoma involves familiarity with the protean patterns and identification, in most cases, of features indicative of invasion and also abnormal cellular features, especially those related to the nucleus. Supportive cytoplasmic and luminal features also play a role. [28, 49, 55]

GLEASON'S GRADING SYSTEM (11, 19, 21, 25, 28, 32, 47, 49, 53, 55, 59)

FIGURE 2



This is the most powerful prognostic indicator of prostate carcinoma.

1. Single, Separate, uniform glands loosely packed, with definite edges.
2. Single, separate, uniform glands loosely packed, with irregular edges.

3A. Single, Separate, uniform glands, scattered.

3B. Single, separate, very small glands, scattered.

4A. Fused glands, raggedly infiltrating.

4B. Same, with large pale cells (hypernephroid)

5A. Almost solid, rounded masses, necrosis ("Comedocarcinoma")

5B. Anaplastic, raggedly infiltrating.

STOUT CRITERIA FOR PROSTATIC ADENOCARCINOMA DIAGNOSIS [11, 53, 55]

1. Glandular pattern-small irregular glands without any particular relation to the adjacent stroma or normal glands.
2. Arrangement of glandular epithelium-lack of basal cells in cancer.
3. Cellular details-large, deeply staining prominent nucleoli.

criteria for diagnosis of prostatic adenocarcinoma. [11, 19, 28, 53, 55]

Major criteria:

Architectural: Infiltrative small glands or cribriform glands .

Single cell layer (absence of basal cells)

Nuclear atypia: Nuclear and nucleolar enlargement.

Minor criteria

- Intraluminal wispy blue mucin
- Pink amorphous secretions
- Mitotic figures
- Intraluminal crystalloids
- Adjacent high grade PIN
- Amphophilic cytoplasm

Specific features of carcinoma

1) Perinerual invasion
2) Collagenous micronodules
3) Glomerulations

PROGNOSTIC FACTORS [49]

Category I - Serum PSA

Gleason grade

Pathologic stage

Surgical margins

Category II - DNA ploidy

Volume of cancer in radical prostatectomy

Volume of cancer in needle biopsies

Histologic subtype

Category III -Perineural invasion

Lymphnode micrometastases

Nuclear roundness

Nuclear chromatin texture

Mitotic figures

MIB-1

PCNA

Apoptosis

PSA derivatives

Androgen receptors

TRANSITIONAL CELL CARCINOMA OF THE PROSTATE:

Most pathologists and urologists have faced the problem of deciding whether a neoplasm is of prostatic or bladder origin. Primary neoplasm of either organ can invade the other. [8, 11 12, 19]

Ende et al postulated that this neoplasm began in the periurethral and prostatic ducts in the area of junction of columnar and transitional epithelium. [11, 19, 25, 28]

Karpas and Moumgius subsequently demonstrated that the hyperplastic transitional cells arise form the reserve cells lying between luminal epithelium and basement membrane in the periurethral ducts. [11, 28, 49]

Johnson et al have atributed one of the three pathophysiological processes for the oncogenesis of transitional cell carcinoma of the prostate;

1) Direct extension from a contiguous infiltrating vesical or prostatic urethral malignant lesions.
2) Prostatic urethral implantation from an epithelial tumour elsewhere.
3) Carcinogenic stimulation of periprostatic glandular urothelium resulting in neoplastic changes in the epithelium which may proceed to an infiltrating prostate lesion. [28,49,55]

PROLIFERATIVE STATUS

Mitotic figures are rarely found in tissue sections in normal or hyperplastic prostatic epithelium.The number of mitotic figures increased progressively from benign epithelium through PIN to cancer. Adenocarcinomas with cribrifrom growth patterns and those composed of solid areas of undifferentiated tumour cells contained most mitotic figures. Therefore, proliferative cell nuclear antigen (PCNA) and Ki-67 antigens are used to assess miitotic activity. [4, 11, 13]

The number of mitotic figures correlated with cancer stage and grade as well as with the progression and progression free survival. Androgen deprivation therapy results in a dramatic decline in the number of mitotic figures in cancer. [4, 13, 19, 34]

In prostate cancer, a high proliferation index for ki-67 appear to add predictive information for patient outcome above the traditional indicators of Gleason score, pathologic stage and DNA ploidy. However ki-67 labelling index may discriminate between organ corfined and metastatic cancer. Hence, elevation in the proliferation indices of Ki-67,MIB-1 appears to reflect progression. In combination , these findings suggest that ki-67 expression may be one of the predictors of recurrence, progression and survival. [4, 7, 11, 13, 28, 56]

In post-radiation reccurence prostatic adenocarcinoma, the sole proliferation index by Ki-67 reached independent significance. Diagnostically this may become an important diagnostic factor as the number of patients surviving long-term following radical radiotherapy increases. [4, 28]

OBSERVATION AND RESULTS

This prospective study included 120 cases of prostatic lesions which included Benign Nodular Hyperplasia,Prostatic abscess,precancerous lesions and frankly malignant lesions of varying grades.The clinical data including age of the patients,presenting symptoms,clinical diagnosis and histopathological data are listed in the master chart.

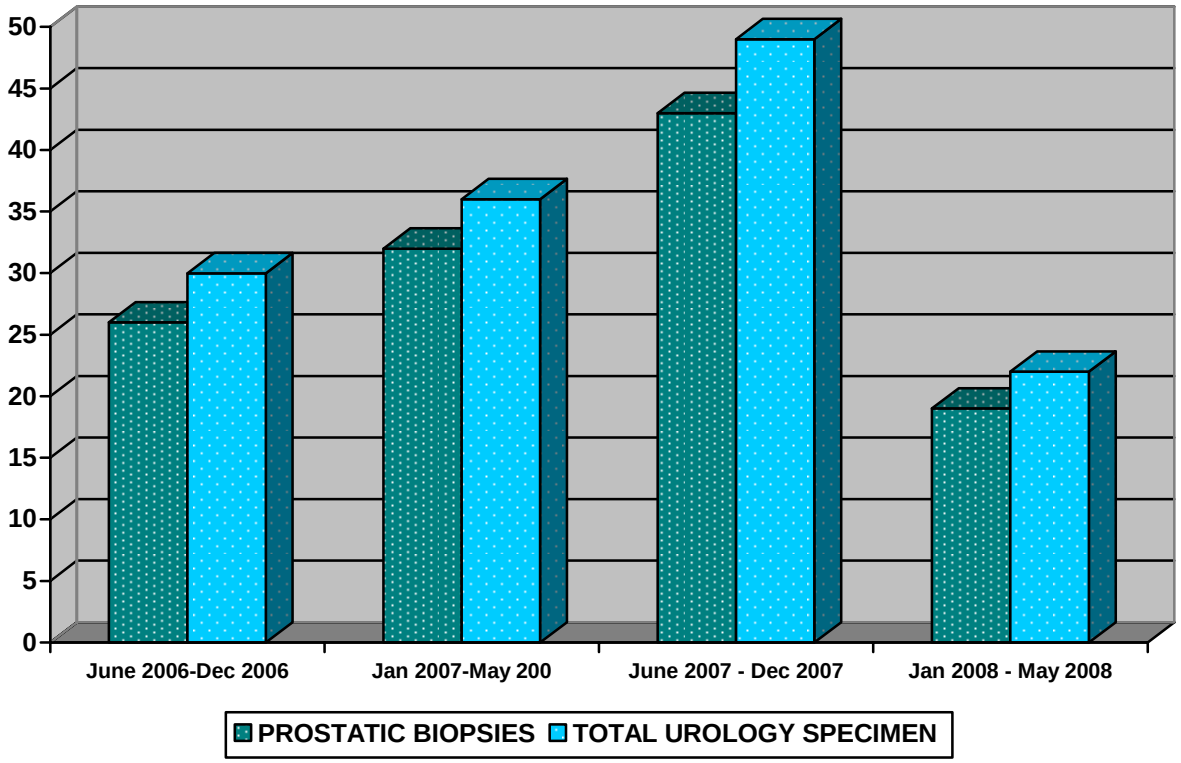
Incidence:

The incidence of prostatic lesions in this study is 1.8% (120 cases) of the total number of general biopsy specimen.This accounted for 87.5% of total urology specimen received during the period of 2006-2008.

TABLE : 1

Period	Prostate biopsies	Total Urology specimen
June ’06-Dec’06	26 cases	30
June ’07-May’07	32 cases	36
June ’07-Dec’07	43 cases	49
June ’08-May’08	19 cases	22
Total	120 cases	137

FIGURE : 3

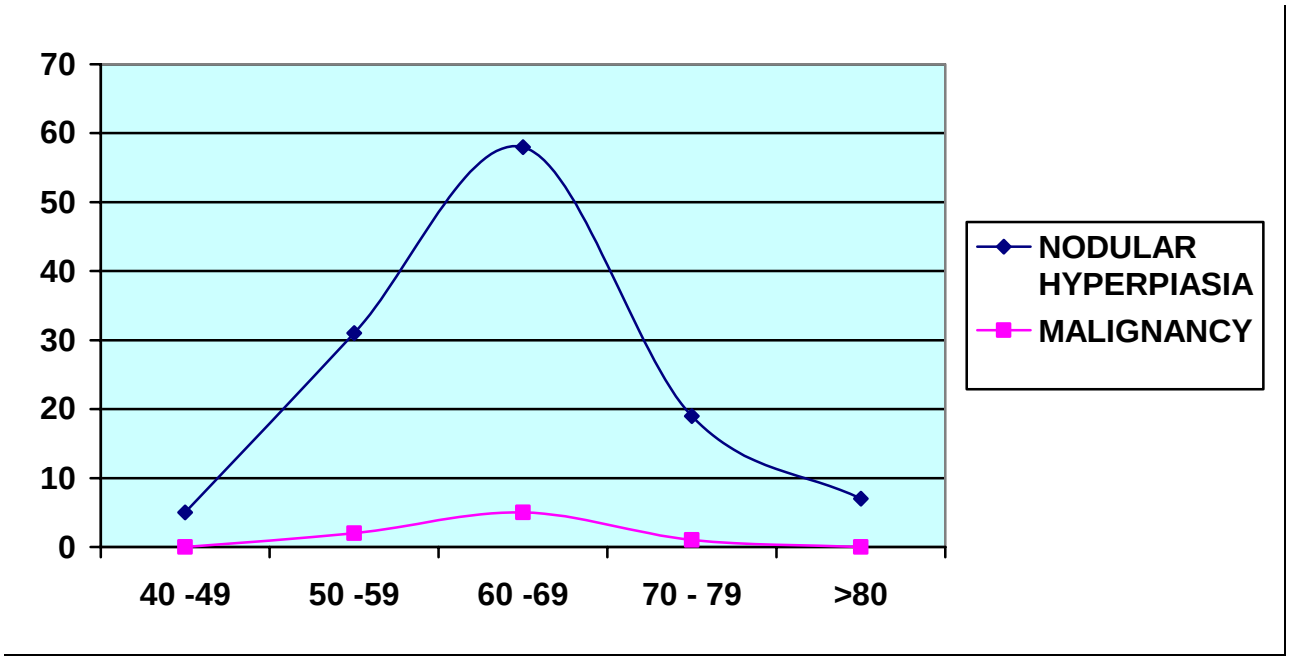


This clearly shows the number of patients presenting with prostatic symptoms in the urology clinic and the considerable morbidity due to prostatic diseases.

Age incidence:

The distribution of cases according to age is shown in the graph.Out of 120 cases studied,maximum number of cases of both Nodular Hyperplasia and malignancy were seen in the seventh decade-49%

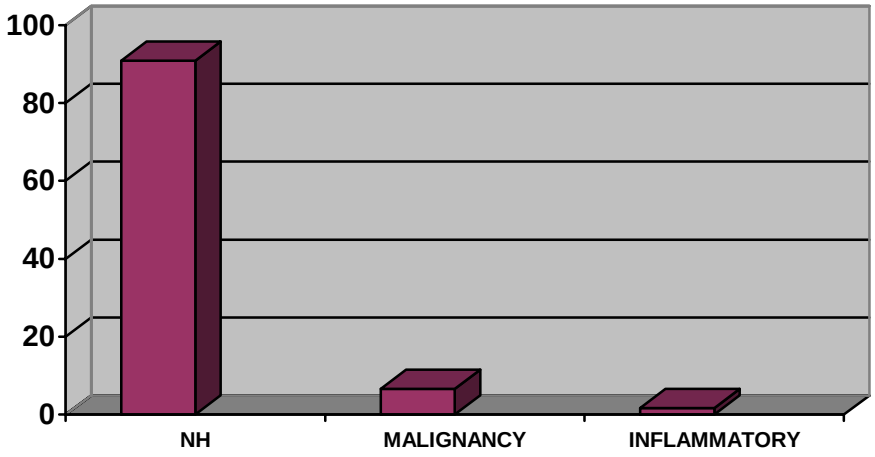
FIGURE : 4



Distribution of prostatic lesions:

The following bar diagram shows the incidence of different prostatic lesions.Benign nodular hyperplasia is by far the most common type of lesion-90.8% (109 cases) followed by carcinoma 6.6 % (8 cases) and inflammatory lesions-1.7% (2 cases).

FIGURE : 5



BENIGN NODULAR HYPERPLASIA

Out of all the prostatic lesions studied, Nodular hyperplasia constituted the bulk of the lesions (90.8%).

The following table shows the distribution of cases of nodular hyperplasia according to the age. The youngest patient reported in this study was 41 years and oldest patient was 83 years old.A progressive increase is observed in the incidence until the 7^{th} decade.

TABLE 2

Age	Number of cases	Percentage
5^{th} decade	5	45%
6^{th} decade	29	25.9%
7^{th} decade	53	47.3%
8^{th} decade	18	16.1%
9^{th} decade	6	5.4%

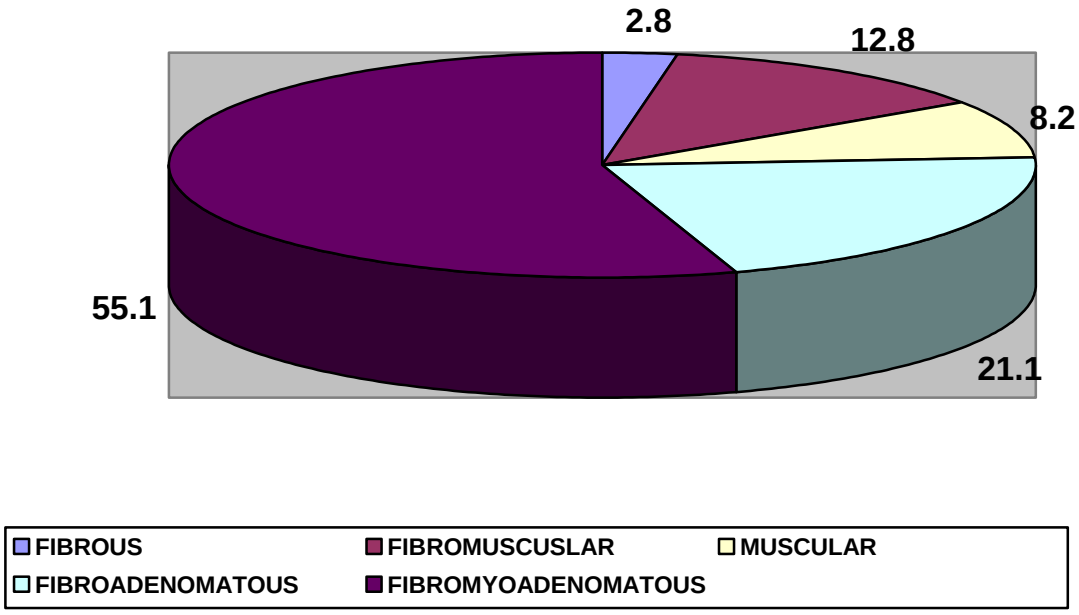
Clinical grading of Nodular Hyperplasia:

The distribution of various degrees of enlargement of the prostate in different age group was studied..In this study, the patients presented with grade II and III only and no single case was noted with I/IV. Grade II –100 Cases, Grade III-9 cases.Similar results were documented by the earlier work carried out by Hunt et al in which 8.7% of cases presented with grade I enlargement and 48% of cases with grade II enlargement.Grade III to IV enlargement of the prostate was observed in 33.9% and 7.8% of cases respectively. There was no significant correlation between the degree of enlargement of prostate and the morphological type of nodule.

Morphological types of nodules:

The incidence of 5 types of nodules is shown in the following pie chart. Fibromyoadenomatous nodule was the most common type encountered (55.1% - 60 cases) followed by fibroadenomatous nodules (21.1%-23 cases). The fibromuscular and muscular nodules constituted 12.8% - 14 cases and 8.2% - 9 cases respectively. The least common type of nodule observed in this study was stromal nodule (2.8% - 3 cases).

FIGURE : 6



Status of basal cells in Nodular Hyperplasia:

All the sections of nodular hyperplasia were screened for the evidence of basal call hyperplasia, Nodular aggregates of basal cells were identified in 42 cases (35%) in that 8 cases had complete basal cell hyperplasia. In 26 cases (23.2%) basal cell hyperplasia was associated with atrophic changes in the acini.

Prostatic intraepithelial Neoplasia (PIN):

All the sections of Nodular Hyperplasia were carefully scrutinized for the evidence of PIN and graded when present. Low grade PIN was noted in 33 cases-27.5% and high grade PIN in 3 cases-25% .

Other associated findings:

In 51 cases (42.5%) focal or diffuse lymphocytic infiltration was present. 5.8% cases (7cases) showed squamous metaplasia and 6.7% (8 cases) showed transitional metaplasia of the acini. Corpora amylacea was noted in 76.8% (86 Cases).

Many cases showed cystic dilatation and flattening of the lining cells atleast focally.

MALIGNANT LESIONS OF THE PROSTATE

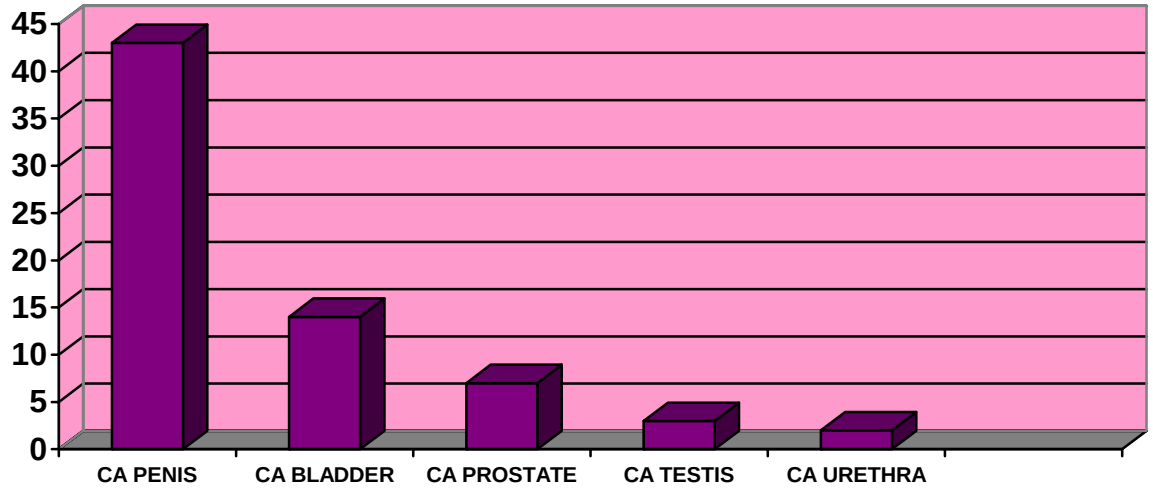
Incidence:

In our prospective study, malignant lesions constituted the second most common pathology of prostate.

This study includes a total of 8 cases (6.6%) of malignant lesions of the prostate.

We also observed that carcinoma of prostate was the third common malignancy of the male genital tract next only to carcinoma of penis and bladder.Prostatic carcinoma was closely followed by malignancy of testis and urethra, which is shown in the following bar diagram.

FIGURE : 7



Age incidence:

The age at diagnosis of prostatic carcinoma reported in this study ranged form 51 to 70 years with average age of 61 years.The age distribution is as follows

- **50-59 year 2 cases 25%**
- **60-69 year 5 cases 62.5%**
- **70-79 year 1 case 12.5%**

Maximum number of cases were seen in the 7^{th} decade.

Histologic distribution of malignant lesions:

8 cases of malignancies of the prostate were studied. Adenocarcinoma was the most common (7 cases-87.5%) type of primary carcinoma encountered. 1 case (12.5%) of transitional cell carcinoma of prostate was noted.

Grading of adenocarcinoma of the prostate:

Gleason's grading system of adenocarcinoma cases is shown in the following table.Based on Gleason's criteria, 7 cases of adenocarcinoma were reviewed and assigned to the specific category according to the five histological patterns.

In the Gleason's, tertiary grading system,among the five patterns described the predominant pattern present in the biopsy was noted and assigned the first number.The second common and the least common patterns were noted and assigned the second and third numbers respectively.Finally all these three numbers were added together and the Gleason's score thus obtained.

GLEASON'S TERTIARY GRADING SYSTEM FOR CARCINOMA:

TABLE : 3

S.No	PATH NO	HPE DIAGNOSIS	GLEASON'S GRADE	GLEASON'S SCORE
1.	3034/06	Adenocarcinoma	2+3+1	6
2.	266/07	Adenocarcinoma	4+5+3	12
3.	1484/07	Adenocarcinoma	5+3+4	12
4.	2350/07	Adenocarcinoma	3+2+1	6
5.	2644/07	Adenocarcinoma	2+3+1	6
6.	1030/08	Adenocarcinoma	2+3+1	6
7.	1092/08	Adenocarcinoma	4+3+2	9

This shows the presence of higher grades(3,4,5) as the predominant pattern in the majority of the cases which will clearly affect the prognosis.

Prostatic intraepthelial neoplasia:

Foci of high grade intraepithelial neoplasia was noted in all the 7 cases (100%) of adenocarcinoma.

Perineural invasion:

Only one case (12.5%) showed the evidence of perineural invasion in the present study whereas a study by Rodis et al reported an incidence of 41.6%.It seems likely that perineural invasion would have been demonstrated in additional cases, if radical prostatectomy specimen and complete serial sections would have been possible.

Mucin histochemistry in prostatic lesions:

15 cases were selected for PAS and alcian blue staining, including 7 cases of frank adenocarcinoma, 1 case of transitional cell carcinoma, 2 cases of high grade PIN, 2 cases of low grade PIN, 1case of atypical adenomatous hyperplasia and 2 cases of Nodular Hyperplasia.

Type of stain	**Purpose**	**Result**
PAS	Stains neutral mucins and glycogen	Glycogen and related mucin-**Pink**
Alcian blue at PH 2.5	Stains all acid mucins	Total acid mucin -**Blue**

The following table shows the results of mucin histochemistry.

MUCIN HISTOCHEMISTRY TABLE:

TABLE : 4

S.No	PATH NO	HPE DIAGNOSIS	ALCIAN BLUE	PAS
1.	342/08	Nodular Hyperplasia	Negative	Positive
2.	560/08	Nodular Hyperplasia	Negative	Positive
3.	3399/07	AtypicalAdenomatousHyperplasia	Positive	Negative
4.	222/08	Nodular Hyperplasia, LGPIN	Negative	Positive
5.	223/08	Nodular Hyperplasia, LGPIN	Negative	Positive
6.	2863/06	Nodular Hyperplasia, HGPIN	Negative	Positive
7.	267/08	Nodular Hyperplasia,HGPIN	Positive	Negative
8.	3034/06	Adenocarcinoma	Negative	Positive
9.	266/07	Adenocarcinoma	Positive	Negative
10.	1484/07	Adenocarcinoma	Positive	Negative
11.	2350/07	Adenocarcinoma	Positive	Positive
12.	2600/07	Adenocarcinoma	Negative	Positive
13.	1030/08	Adenocarcinoma	Negative	Positive
14.	1092/08	Adenocarcinoma	Positive	Negative
15.	3411/07	Transitional Cell Carcinoma	Negative	Negative

Results:

- In 4 cases (57.1%) of adenocarcinoma, intraluminal blue stained acidic mucin could be demonstrated.
- All cases of Nodular Hyperplasia and LGPIN were devoid of acidic mucin.
- 1 out of 2 cases of HGPIN showed the presence of acid mucin.
- 1 case of AAH showed positivity for acidic mucin. The intensity of staining was similar in both carcinoma and AAH. These findings indicate that there is a close relation in mucin expression between AAH and well differentiated adenocarcinoma.

The incidence of acidic mucin was not only higher in prostatic carcinomas but also directly proportional to the tumour grade in all but the poorly differentiated ones..

Transitional cell carcinoma was negative for both acidic and neutral mucin.

Immunohistochemistry with Ki-67:

10 cases were selected to study the status of proliferative activity which included 2 cases of nodular hyperplasia, 2 cases of LGPIN, 2 cases of HGPIN and 4 cases of carcinoma of varying grades.

The following table shows the Ki-67 Labelling Index (Ki-67 LI)obtained in each case;

TABLE :5

S.No	Path no	HPE Diagnosis	Ki-67 LI
1	266/08	Nodular Hyperplasia	0
2	562/08	Nodular Hyperplasia	1
3	3108/06	LGPIN	1
4	2567/07	LGPIN	2
5	2863/06	HGPIN	3
6	267/08	HGPIN	3
7	3034/06	Low Grade Ca	9
8	2644/07	Low Grade Ca	10
9	2350/07	High Grade Ca	10
10	1484/07	High Grade Ca	27

Results:

- Both cases of Nodular Hyperplasia showed less than 1% positive staining. One case of low grade PIN should 1% and the other showed 2% as Ki-67 labelling index.
- Both the two cases of high grade PIN showed 3% as ki-67 labelling index.
- Among carcinoma, one case with Gleason's score of 6 showed 9% Ki-67 LI, other with score 6 showed 10% Ki-67 LI. Two higher grade carcinomas with scores 9 and 12 had 12% and 27% as their Ki-67 LI respectively.

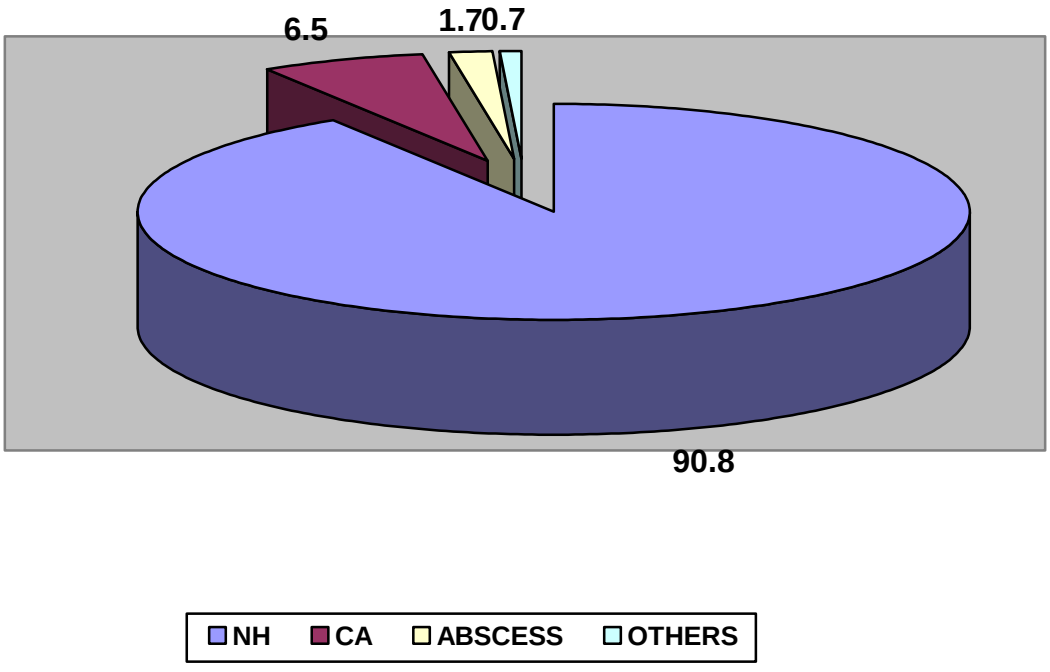
Thus, the proilferative status increased progressively from benign epithelium through PIN to cancer. Even in carcinoma higher grade carcinomas had higher labelling index than lower grade carcinomas.

DISCUSSION

Prostate hosts a number of diseases ranging from inflammation to carcinoma.This leads to considerable morbidity and mortality worldwide.

Among the diseases of prostate, benign nodular hyperplasia is the most commonly observed pathology. In our Prospective study, the distribution of prostatic lesions is shown in the following pie chart.

FIGURE : 8



BENIGN NODULAR HYPERPLASIA

The development of nodular hyperplasia is age related.[11,19,20] In our study,the youngest patient was 40 years old and the oldest patient was 82 years old.Many pathologists have proved that testosterone is a powerful stimulation for the development of hyperplasia. This is also supported by the regression of the nodules after androgen deprivation [32,47,48,49]

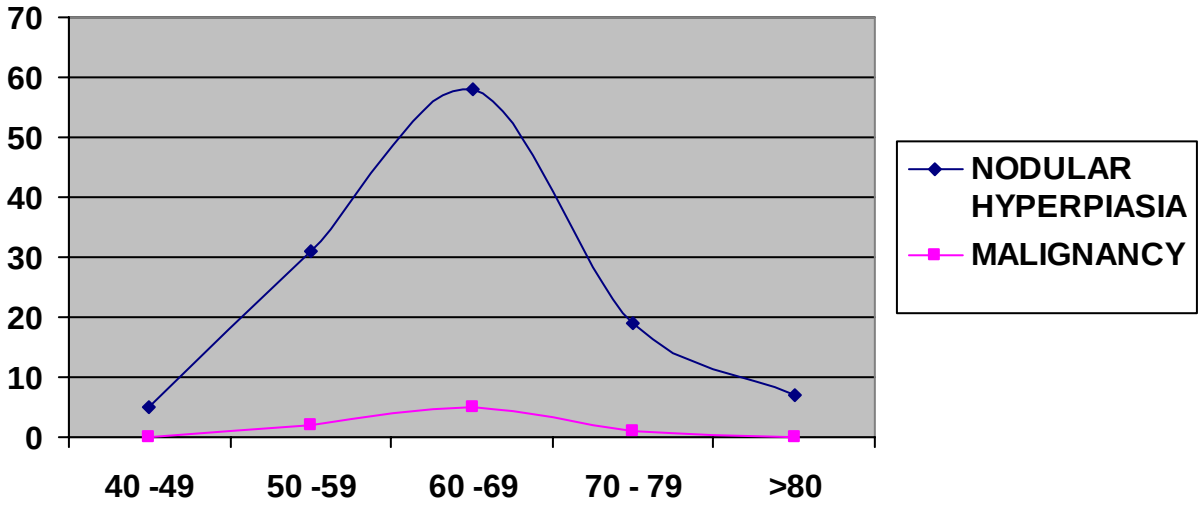
The principal androgenic hormones are testosterone and dihydrotestosterone (DHT).The DHT level is elevated in tissues with nodular hyperplasia compared to normal tissue in some studies There is no clear relationship between plasma DHT levels and the clinical presence of nodular hyperplasia [41,47,48,49]

Current experimental work is looking at the exact roles of hormone growth factors and their respective receptors.

There is also considerable interest in the role of programmed cell death in the development of nodular hyperplasia.[28,32,44]

In our prospective study,the highest incidence of nodular hyperplasia was noted in the 7^{th} decade. The number of cases in each decade in shown in the following graph .

FIGURE : 9



Age incidence of nodular hyperplasia-A comparative analysis.

TABLE : 6

S.No	STUDY	TOTAL NO OF CASES	40-49 Yrs,%	50-59 Yrs,%	60-69 Yrs,%	70-79 Yrs,%	>80 Yrs,%
1.	YOUNG	450	14, 3.1%	79, 17.6%	199, 44.2%	146, 34.4%	12, 2.7%
2.	HUNT	1000	16, 1.6%	258, 25.8%	564, 50.4%	155, 15.5%	7, 7. 7%
3.	KARLE BEECHAAM	60	6, 10%	14, 23.3%	30, 50%	9, 15%	1, 1. 7%
4.	KRETSCHMER	651	13, 2%	130, 19.9%	311, 47.7%	166, 25.5%	31, 4.8%
5.	PRESENT STUDY	112	5, 4.5%	29, 25.9%	53, 47.3%	18, 16.1%	6, 5.4%

The incidence of nodular hyperplasia in each decade observed in our study is comparable with other similar studies.

The actual incidence of Nodular Hyperplasia should be more in 8th and 9th decades.But, due to age related deaths, the incidence in spuriously lesser.

Regarding morphological subtype of benign nodular hyperplasia , Frank classified it in to 5 types and our study states that fibromyoglandular is the commonest subtype observed.

Basal cell hyperplasia

Since basal cell hyperplasia is usually seen in cases of nodular hyperplasia, the clinical features are essentially the same as those of that condition.The only different significant clinical association is with anti androgen therapy. Basal cell hyperplasia is typically found in the transition zone and is therefore usually identified in the transurethral resection.This may also be atrophy associated and found in the peripheral zone. [11,28,32,55]

In our study,42 cases of nodular hyperplasia were associated with basal cell hyperplasia.In them,8 cases had complete basal cell hyperplasia.In 26 cases of nodular hyperplasia, basal cell hyperplasia was associated with atrophic changes in the acini.This correlates well with previous studies by Young and Frankel et al.

PIN :

Prostatic intraepithelial neoplasia is best characterized as a neoplastic transformation of the 1ining epithelium of prostatic ducts and acini. By definition,this process is confined to within the epithelium, therefore intraepithelial.[45,53]

There is limited literature characterizing the epidemiology of high grade prostatic intraepithelial neoplasia. Based on few recent autopsy studies that included HGPIN in their analysis, it appears that similar to prostate cancer, HGPIN can be detected microscopically in young males, its prevalence increased with age and HGPIN shows strong association with cancer in terms of coincidence in the same gland and its spatial distribution.[45,48,49]

Sakr et al. identified HGPIN in 7,26,46,72,75 and 91% of African-Americans between the third and eighth decades compared to 8,23,29,49,53, and 67% for caucasian men.[59] The incidence of HGPIN in transurethral resection of the prostate is relatively uncommon with two studies reporting a rate of 2.3 and 2.8 % respectively.[28]

Our study correlates well with this, stating 2.5% prevalence of HGPIN in TURP specimen.

The prevalence of high grade PIN in radical prostatectomy or TURP for carcinoma is remarkably high reflecting the strong association with malignancy. Investigators have found HGPIN in 85-100% of radical prostatectomy specimen.[28]

Our study included 7 cases of prostatic adenocarcinoma, which were all TURP specimen.All cases exhibited atleast focal areas of HGPIN.

We also observed that the association of LGPIN with nodular hyperplasia was 27.5%

There are many evidences to suggest morphological relationship of HGPIN and prostatic adenocarcinoma:

- The incidence and extent of both lesions increase with patient age.
- There is an increased frequency,severity and extent of HGPIN in prostatic carcinoma.
- Both HGPIN and carcinoma are multifocal with a predominant peripheral zone distribution.
- Histological transition from HGPIN to cancer has been described.
- HGPIN is more strongly associated with intermediate and high grade prostatic carcinoma.
- There is significant correlation between volume of HGPIN and the number of lymphnode metastases. [28,49,55,59]

Risk of subsequent cancer detection after rebiopsy [28]

Needle biopsy diagnosis % of carcinoma on rebiopsy

- Benign prostatic tissue 20%
- 2) High grade PIN 30%
- 3) PIN with small atypical glands 53%

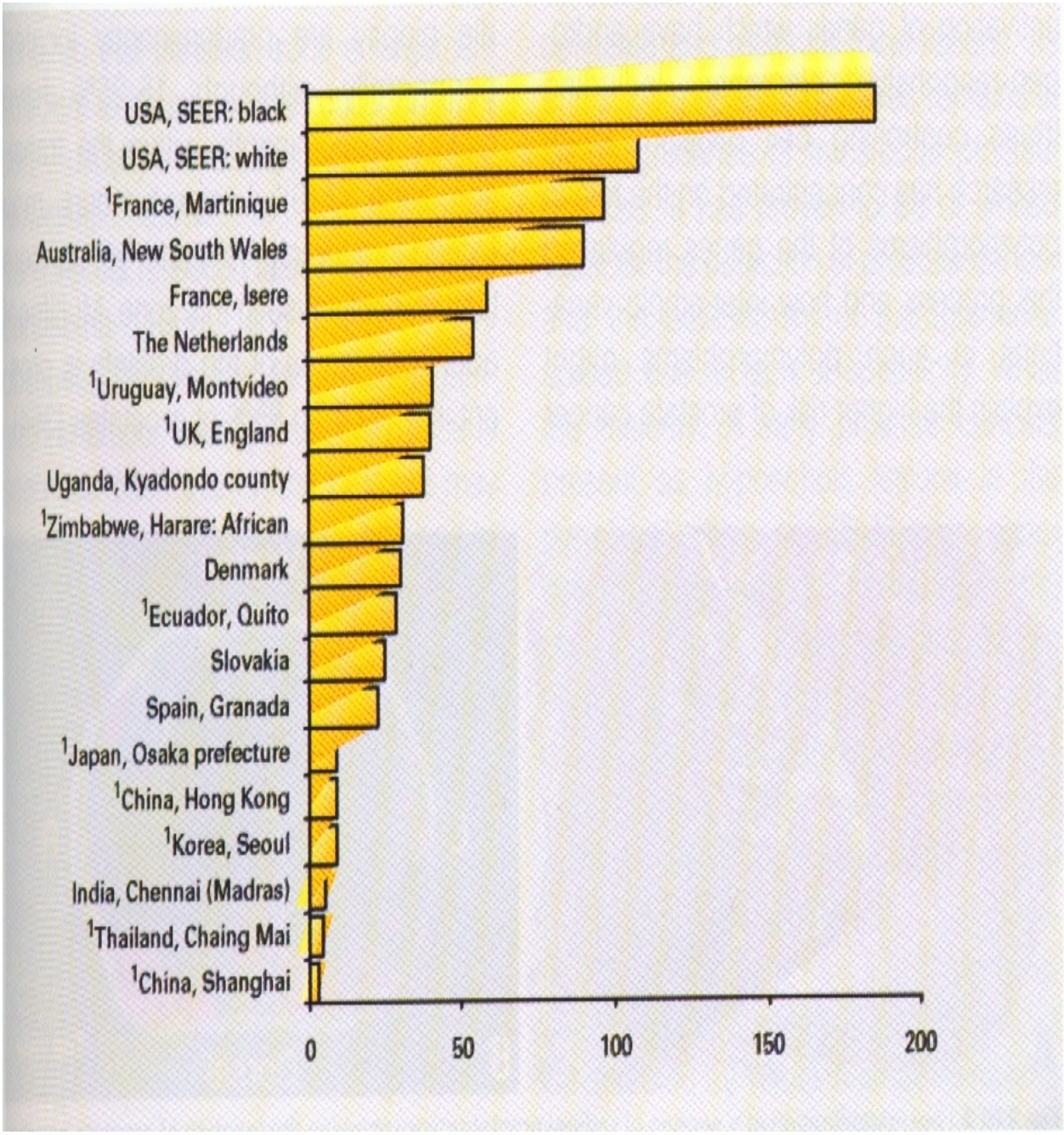
Prostatic adenocarcinoma:

Prostatic cancer contributes significantly to the over all cancer burden, being the most frequent malignancy in men,worldwide.The number of cases had continuosly increased over the past decades,partly due to the higher life expectancy.An additional factor is the western lifestyle characterized by a high caloric diet and lack of physical exercise.Epidemiological data suggest that black people are most susceptible,followed by white people,while the Asians have the lowest risk.[28,32,41,47,53,55]

Histopathological diagnosis and grading play a major role in the management of prostate cancer.[28,47,53]

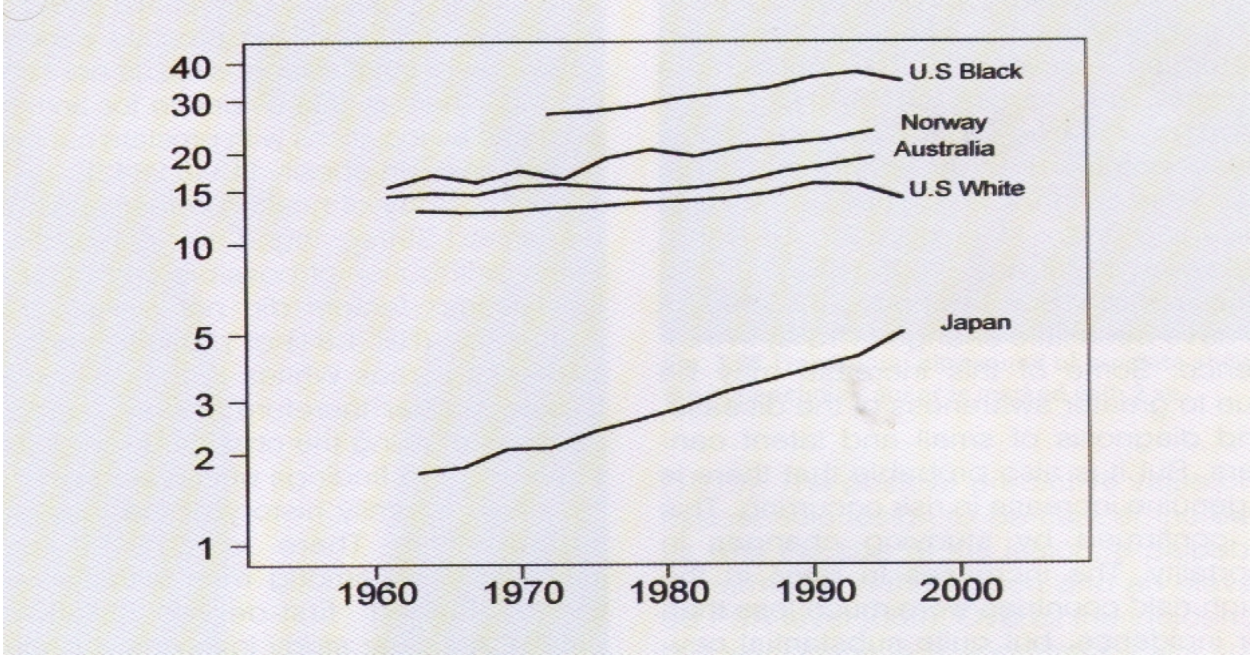
PROSTATE CANCER INCIDENCE:ASR/10

FIGURE : 10



International trends in age standardized mortality rates of prostate caner(World Standard) source:WHO/NCHS

FIGURE : 11



The risk of prostate cancer rises very steeply with age.Incidence of clinical disease is low until after age 50,and then increases at approximately the 9-10 th power of age,compared with the 5-6 th power for other epithelial cancers.Worldwide,about three-quarters of all cases occur in men aged 65 or more.[28,47,53]

Our study includes 8 cases of prostatic carcinomas which account for 5.8% of total urology specimen and 6.6% of prostatic biopsies.The age of the patients ranged from 51-70 years with an average of 61 years.

The distribution of cases according to the age is shown below:

50-59 year	**2 cases**	**25%**
60-69 year	**5 cases**	**62.5%**
70-79 year	**1 case**	**12%**

The youngest patient was 51 years old and the oldest patient was 70 years old.

Features helpful in the diagnosis of adenocarcinoma in needle biopsy specimen [49]

1) Infiltrative pattern

2) Nuclear enlargement

3) Prominent nucleoli

4) Pink amorphous secretions

5) Amphophilic cytoplasm

6) Blue tinged mucinous secretions

7) Nuclear hyperchromasia

8) Intraluminal crystalloids

9) Mitotic figures

10) High grade PIN

Features specific for malignancy:

- Glomerulations
- Mucinous fibroplasia
- Perineural invasion

In our study, the features commonly noted were infiltrative pattern,nuclear enlargement,prominent nucleoli,adjacent HGPIN,amphophilic cytoplasm and mitotic figures.

All cases were associated with adjacent HGPIN in our study and in one case we noted perineural invasion which is a special feature of prostatic carcinoma.We have not observed glomerulations or collagenous micronodules.

GLEASON'S GRADING SYSTEM:

This is the most powerful prognostic indicator of prostatic adenocarcinoma.[19,28,53,55] We applied Gleason's tertiary grading system for all adenocarcinomas and the following table shows the Gleason's grade and score of the 7 carcinomas

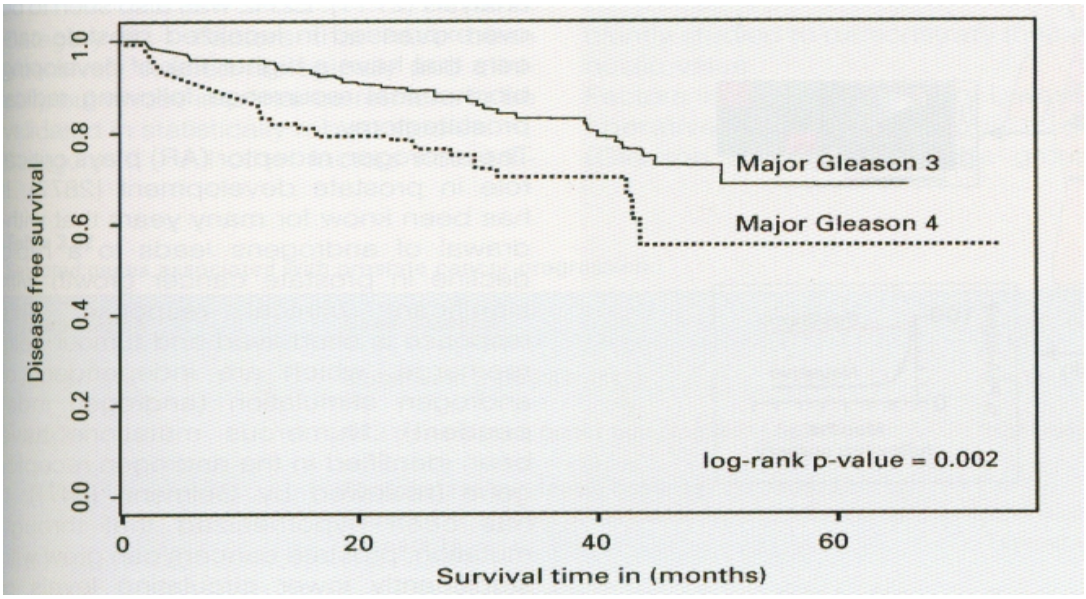
GLEASON'S TERTIARY GRADING SYSTEM FOR CARCINOMA:

TABLE : 7

S.No	PATH NO	HPE DIAGNOSIS	GLEASON'S GRADE	GLEASON'S SCORE
8.	3034/06	Adenocarcinoma	2+3+1	6
9.	266/07	Adenocarcinoma	4+5+3	12
10	1484/07	Adenocarcinoma	5+3+4	12
11	2350/07	Adenocarcinoma	3+2+1	6
12	2644/07	Adenocarcinoma	2+3+1	6
13	1030/08	Adenocarcinoma	2+3+1	6
14	1092/08	Adenocarcinoma	4+3+2	9

The higher the grade,the less favourable is the prognosis and this is clearly shown in the following figure.

FIGURE : 12

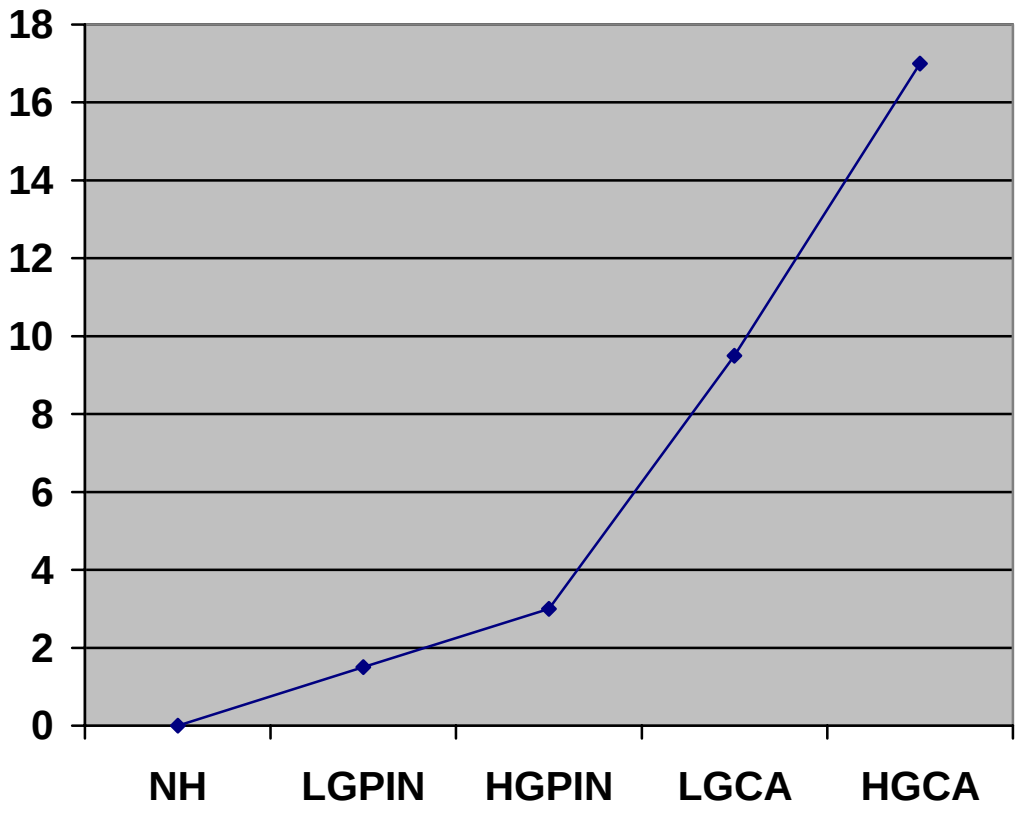


Ki-67 LABELLING INDEX (Ki -67 LI) :

Many recent literature clearly document the importance of proliferative markers in assessing the prognosis of prostatic carcinoma.[4,13,33]

The mitotic activity gradually increased form benign epithelium through PIN to malignancy. We observed the same in our study with **ki-67** marker.We selected 10 cases which included Nodular hyperplasia,LGPIN, HGPIN, low grade and high grade carcinomas. The results are plotted in the graph taking the average of each of the two similar cases,and the graph goes fairly well with the literature.

FIGURE : 13

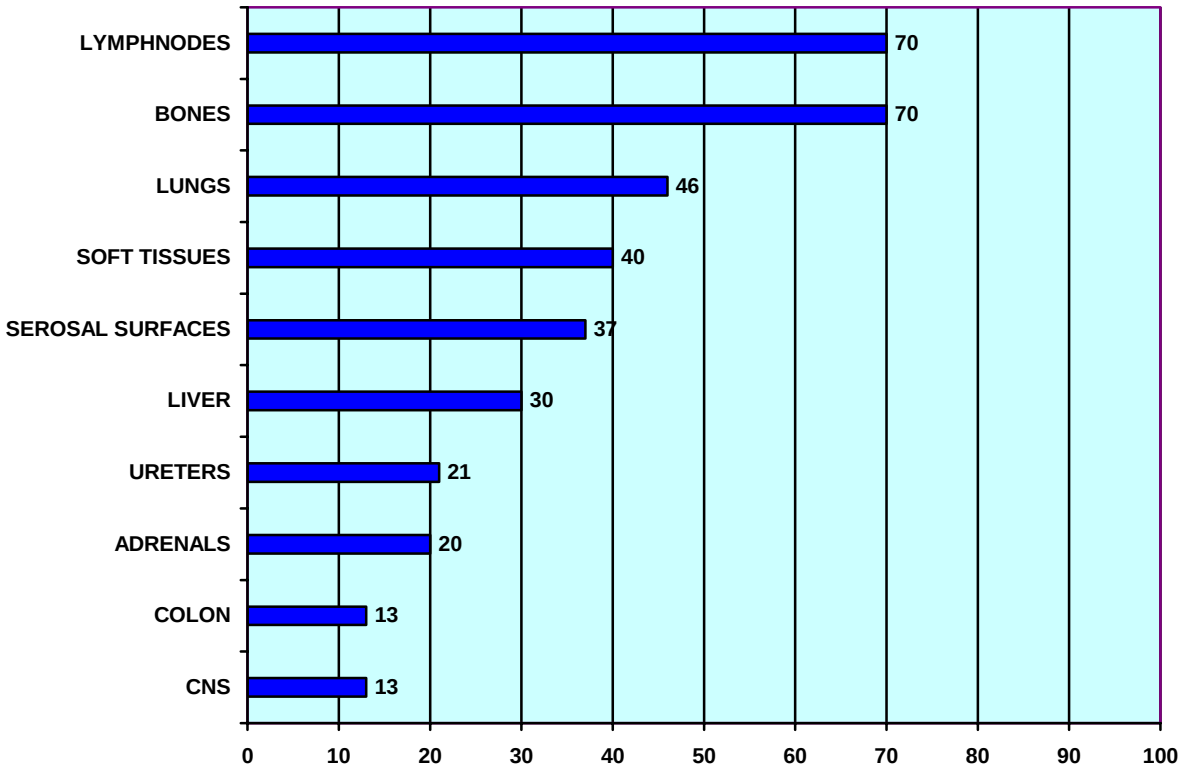


Metastasis:

Prostatic carcinoma is well known for its local spread and distant metastases. The common sites of local spread are periprostatic tissues,seminal vesicle,urethra and bladder. The most common sites of prostate cancer metastases is shown in the following figure.[28,47,53,55])

THE TEN MOST COMMON SITES OF PROSTATE CANCER METASTASES AT AUTOPSY IN THE UNITED STATES AND JAPAN

FIGURE : 14



In our study, we lost the patients for follow up and therefore we could not get the incidence and common sites of metastases.

CONCLUSION

In the present prospective study comprising of 120 cases of prostatic lesions evaluated with clinical,light microscopy,histochemical and IHC, following conclusions are made and presented.

1) Benign Nodular Hyperplasia is the most commonly observed pathology of prostate.
2) The age incidence of Nodular Hyperplasia is between 5 th and 7 th decade.
3) Fibromyoadenomatous nodule is the most frequently encountered type of Nodular Hyperplasia.
4) The incidence of Low Grade Prostatic Intraepithelial Neoplasia is 27.5% and High Grade Prostatic Intraepithelial Neoplasia is 2.5 % in TURP specimen of Nodular Hyperplasia.
5) Among the primary malignant neoplasms of the prostate,Adenocarcinoma is the commonest.
6) All cases of Adenocarcinoma showed atleast focal presence of adjacent High Grade Prostatic Intraepithelial Neoplasia.
7) Higher grades, according to Gleason's Grading system are more commonly observed as the predominant pattern,rather than lower grades.
8) Normal and hyperplastic glands exhibited the presence of neutral mucin whereas,High Grade Prostatic Intraepithelial Neoplasia,Atypical Adenomatous Hyperplasia and carcinomas showed predominance of acidic mucin.
9) Mitotic activity progressively increased from benign epithelium through Prostatic Intrraepithelial Neoplasia to malignancy.

10)Even among malignant lesions,those with higher Gleason's grade and scores had higher Ki-67 labelling index than those with lower Gleason's grade and score.

KI-67 LABELLING INDEX which was widely used in various other neoplasms,has now gained importance as an indicator of cell proliferation in prostatic carcinoma.In normal and benign epithelium,mitotic activity is indiscernible.In rapidly proliferating epithelium such as in precursor lesions and prostatic carcinoma, mitotic activity increases steadily and it can be readily shown by Ki-67 Labelling Index.

So,**ASSESSING THE MITOTIC ACTIVITY WITH A PROLIFERATIVE MARKER SUCH AS KI-67 IS A REPRODUCIBLE, SIMPLE,EFFICIENT, RELIABLE AND RELATIVELY INEXPENSIVE METHOD AND CAN BE USED AS AN ADJUNCT TO ROUTINE DIAGNOSIS OF PROSTATIC CARCINOMA ESPECIALLY IN DOUBTFUL CASES AND ALSO IN ASSESSING THE PROGNOSIS**.

MASTER CHART

S.NO:	PATH NO	AGE	CLINICAL DIAGNOSIS	HPE DIAGNOSIS
1	1719/06	45	BPH – GRADE II	NODULAR HYPERPLASIA
2	1758/06	50	BPH – GRADE II	NODULAR HYPERPLASIA
3	1759/06	68	BPH – GRADE II	NODULAR HYPERPLASIA
4	1820/06	60	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
5	1821/06	72	BPH – GRADE II	NODULAR HYPERPLASIA
6	1852/06	56	BPH – GRADE II	NODULAR HYPERPLASIA
7	2107/06	70	BPH – GRADE II	NODULAR HYPERPLASIA
8	2124/06	60	BPH – GRADE II	NODULAR HYPERPLASIA
9	2125/06	58	BPH – GRADE II	NODULAR HYPERPLASIA
10	2623/06	75	BPH – GRADE II	NODULAR HYPERPLASIA
11	2863/06	64	BPH – GRADE II	NODULAR HYPERPLASIA,HGPIN
12	3034/06	60	BPH – GRADE II	ADENOCARCINOMA
13	3038/06	70	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
14	3086/06	80	BPH – GRADE II	NODULAR HYPERPLASIA
15	3163/06	63	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
16	3241/06	60	BPH – GRADE II	NODULAR HYPERPLASIA
17	3242/06	65	BPH – GRADE II	NODULAR HYPERPLASIA
18	3247/06	60	? CARCINOMA	CLUSTER OF DYSPLASTIC CELLS
19	3290/06	75	BPH – GRADE III	NODULAR HYPERPLASIA
20	3292/06	60	BPH – GRADE II	NODULAR HYPERPLASIA
21	3312/06	50	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
22	3384/06	56	BPH – GRADE II	NODULAR HYPERPLASIA
23	3433/06	60	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
24	3444/06	62	BPH – GRADE II	NODULAR HYPERPLASIA
25	3496/06	70	BPH – GRADE II	NODULAR HYPERPLASIA
26	3497/06	55	BPH – GRADE II	NODULAR HYPERPLASIA
27	266/07	51	BPH – GRADE II	ADENOCARCINOMA
28	268/07	55	BPH – GRADE II	NODULAR HYPERPLASIA
29	339/07	63	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
30	340/07	55	BPH – GRADE II	NODULAR HYPERPLASIA
31	362/07	61	BPH – GRADE II	NODULAR HYPERPLASIA,HGPIN
32	443/07	65	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
33	531/07	40	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
34	892/07	60	BPH – GRADE II	NODULAR HYPERPLASIA
35	933/07	70	BPH – GRADE II	NODULAR HYPERPLASIA
36	934/07	50	BPH – GRADE II	NODULAR HYPERPLASIA
37	960/07	55	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
38	1092/07	65	BPH – GRADE II	NODULAR HYPERPLASIA
39	1094/07	60	BPH – GRADE II	NODULAR HYPERPLASIA

40	1137/07	75	BPH – GRADE II	NODULAR HYPERPLASIA
41	1215/07	60	BPH – GRADE II	NODULAR HYPERPLASIA
42	1250/07	60	BPH – GRADE II	NODULAR HYPERPLASIA
43	1251/07	45	BPH – GRADE II	NODULAR HYPERPLASIA
44	1291/07	82	BPH – GRADE II	NODULAR HYPERPLASIA
45	1292/07	80	BPH – GRADE II	NODULAR HYPERPLASIA
46	1293/07	68	BPH – GRADE II	NODULAR HYPERPLASIA
47	1322/07	65	BPH – GRADE II	NODULAR HYPERPLASIA
48	1323/07	71	BPH – GRADE II	NODULAR HYPERPLASIA
49	1436/07	69	BPH – GRADE II	NODULAR HYPERPLASIA
50	1483/07	60	BPH – GRADE II	NODULAR HYPERPLASIA
51	1484/07	65	BPH – GRADE II	ADENOCARCINOMA
52	1487/07	58	BPH – GRADE II	NODULAR HYPERPLASIA
53	1504/07	60	BPH – GRADE II	NODULAR HYPERPLASIA
54	1505/07	68	BPH – GRADE II	NODULAR HYPERPLASIA
55	1630/07	62	BPH – GRADE II	NODULAR HYPERPLASIA
56	1631/07	60	BPH – GRADE II	NODULAR HYPERPLASIA
57	1726/07	55	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
58	1860/07	64	BPH – GRADE II	NODULAR HYPERPLASIA
59	1867/07	60	BPH – GRADE III	NODULAR HYPERPLASIA,HGPIN
60	1944/07	83	BPH – GRADE II	NODULAR HYPERPLASIA
61	2031/07	60	BPH – GRADE II	NODULAR HYPERPLASIA
62	2092/07	81	BPH – GRADE II	NODULAR HYPERPLASIA
63	2093/07	81	BPH – GRADE II	NODULAR HYPERPLASIA
64	2349/07	55	BPH – GRADE II	NODULAR HYPERPLASIA
65	2350/07	56	BPH – GRADE II	ADENOCARCINOMA
66	2389/07	56	BPH – GRADE II	NODULAR HYPERPLASIA
67	2440/07	60	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
68	2539/07	65	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
69	2567/07	58	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
70	2586/07	45	BPH – GRADE II	NODULAR HYPERPLASIA
71	2635/07	61	BPH – GRADE II	NODULAR HYPERPLASIA
72	2644/07	62	BPH – GRADE II	ADENOCARCINOMA
73	2645/07	51	BPH – GRADE II	NODULAR HYPERPLASIA
74	2646/07	62	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
75	2647/07	45	BPH – GRADE II	NODULAR HYPERPLASIA
76	2689/07	54	ABSCESS	ABSCESS
77	2694/07	57	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
78	2719/07	60	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
79	2802/07	57	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
80	2809/07	50	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN

81	2875/07	71	BPH – GRADE II	NODULAR HYPERPLASIA
82	2876/07	72	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
83	2982/07	72	BPH – GRADE II	NODULAR HYPERPLASIA
84	3003/07	55	BPH – GRADE II	NODULAR HYPERPLASIA
85	3012/07	62	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
86	3044/07	70	BPH – GRADE II	NODULAR HYPERPLASIA
87	3094/07	80	BPH – GRADE II	NODULAR HYPERPLASIA
88	3095/07	63	BPH – GRADE II	NODULAR HYPERPLASIA
89	3108/07	57	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
90	3109/07	65	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
91	3117/07	60	BPH – GRADE II	NODULAR HYPERPLASIA
92	3162/07	70	BPH – GRADE II	NODULAR HYPERPLASIA
93	3216/07	61	BPH – GRADE II	NODULAR HYPERPLASIA
94	3217/07	60	BPH – GRADE II	NODULAR HYPERPLASIA
95	3222/07	60	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
96	3378/07	56	BPH – GRADE II	NODULAR HYPERPLASIA
97	3399/07	75	BPH – GRADE II	ATYPICAL ADENOMATOUS HYPERPLASIA
98	3411/07	53	BPH – GRADE II	TRANSITIONAL CELL CARCINOMA
99	3413/07	60	BPH – GRADE II	NODULAR HYPERPLASIA
100	3429/07	72	BPH – GRADE II	NODULAR HYPERPLASIA
101	3430/07	55	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
102	6/08	60	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
103	67/08	65	BPH – GRADE II	NODULAR HYPERPLASIA
104	138/08	60	BPH – GRADE II	NODULAR HYPERPLASIA
105	151/08	63	BPH – GRADE II	NODULAR HYPERPLASIA
106	222/08	65	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
107	223/08	55	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
108	266/08	65	BPH – GRADE II	NODULAR HYPERPLASIA
109	267/08	70	BPH – GRADE II	NODULAR HYPERPLASIA,HGPIN
110	325/08	59	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
111	340/08	57	BPH – GRADE II	NODULAR HYPERPLASIA
112	341/08	75	BPH – GRADE II	NODULAR HYPERPLASIA
113	342/08	58	BPH – GRADE II	NODULAR HYPERPLASIA
114	560/08	60	BPH – GRADE II	NODULAR HYPERPLASIA
115	561/08	50	BPH – GRADE II	NODULAR HYPERPLASIA,LGPIN
116	562/08	60	BPH – GRADE II	NODULAR HYPERPLASIA
117	878/08	60	BPH – GRADE II	NODULAR HYPERPLASIA
118	939/08	60	BPH – GRADE II	ABSCESS
119	1030/08	70	BPH – GRADE II	ADENOCARCINOMA
120	1092/08	60	CA PROSTATE	ADENOCARCINOMA

APPENDIX

I. HAEMATOXYLIN AND EOSIN

Preparation of the solution:

Distilled water - 1000 ml
Ammonium alum - 100 g
Haematoxylin - 5 g
Absolute Ethyl Alcohol - 50 ml
Mercuric Oxide - 2.5 g

100 g of ammonium alum dissolved in 1000 ml of distilled water by heating and shaking at 60^{o}C.Add solution of 50 g of haematoxylin in 50 ml of ethyl alcohol and bring rapidly to boil.When it begins to boil,remove from flame and add 2.5 g of Mercuric oxide.Mix by swirling gently.

EOSIN STAIN

Eosin Y - 1 g
Distilled water - 20 ml
95% Ethanol - 80 ml
Glacial acetic acid - 0.2ml

Dissolve 1 g eosin Y in 20 ml of water and add 95% ethanol and glacial acetic acid.

Procedure:

1. Sections to water
2. Alum Hematoxylin (Harris)
3. Quick rinse in water
4. Dehydration- 1% acid alcohol- 3 to 4 times
5. Rinse in tap water-30 seconds
6. Stain 1 % aqueous eosin Y for ¼ to 2 minutes
7. Wash in water
8. Clear and mount

APPENDIX

II. PER-IODIC ACID SCHIFF STAINING

Preparation of solution:

Coleman's Feulgen reagent:

Basic Fuchsin	- 1.0 gm
Potassium metasulphite	- 2.0 gm
Normal Hydrochloric acid	- 10 cc
Distilled water	- 200 cc
Activated carbon	- 0.5 gm

Dissolve 1.0 gm Basic Fuschin in 200 cc of hot distilled water.Bring to boiling point.Cool and add 2.0 gm potassium metabisulphite and 10 cc normal hydrochloric acid.Let bleach for 24 hrs.

Add 0.5 gm of activated carbon,shake 1 mt and filter until colourless, store in refrigerator.

1% Per-iodic Acid solution:

Per-iodic Acid crystals	- 1 gm
Distilled water	- 100 cc

Normal Hydrochloric Acid solution:

Hydrochloric Acid Conc sp.Gr 1.19	-83.5 cc
Distilled water	-916.5 cc

Procedure:

1. Smear fixed in methanol for 20 mts
2. Air dried
3. Per-iodic acid for 10 mts(oxidant)
4. Rinse in distilled water
5.Place in Coleman's Feulgen reagent (Schiff's reagent) for 30 mts
6.Wash in tap water for 10 mts for pink colour to develop
7. Immerse in hematoxylin for 1 to 2 mts
8. Rinse in tap water

9. Differentiate in acid alcohol- 2 dips
10. Wash in water 30 sec
11.Two dips in ammonia water 2 %
12. Wash in tap water- 30 sec
13.Dehydrate in absolute alcohol
14.Clear with xylene.

APPENDIX

III- ALCIAN BLUE STAINING

Preparation of solution:

Alcian Blue solution:

Alcian Blue	- 8 gms
Distilled water	- 97 ml
Glacial Acetic Acid	- 3 ml
Thymol	- 1 crystal

Procedure:

1. Sections hydrated in tap water.
2. Placed in 3 % acetic acid for 2 minutes.
3. Stained with Alcian blue solution for 30 minutes.
4. Washed in running tap water for 2 minutes.
5. Rinsed in distilled water.
6. Dehydrated in absolute alcohol.
7. Clearing done with Xylene.

APPENDIX

IV - IMMUNO HISTOCHEMISTRY

Procedure

1. 5 microns thick sections were cut from the blocks received on slides coated with chrome alum gelatin.
2. Slides were dewaxed and dehydrated in graded alcohol.
3. Slides were immersed in 0.3 % Hydrogen peroxide for 20 minutes to block endogenous peroxidase activity.
4. Washed in phosphate buffered saline (PBS)
5. Incubated in primary antibody & Ki – 67.
6. Washed in PBS.
7. Biotinylated link was applied for 20 minutes.
8. Washed in PBS.
9. Incubated in streptavidin-biotin complex.
10. Washed in PBS
11. DAB was used as chromogen.
12. Washed and can be stained with haematoxylin.
13. Mounted with coverslip.

BIBLIOGRAPHY

1. A.Jan, C.Liao, Z.Mo and Y.cao- Meta-analysis of holmium laser enucleation VS transurethral resection of the prostate for symptomatic prostatic obstruction; British journal of surgery; Vol-94; October 2007; 1201-1208
2. Adebaye O.Osunkoya, George J.Metto, Jonathan I.Epstein – colorectal adenocarcinoma involving the prostate, Report of 9 cases, Human Pathology (2007) 38; 1836 -1841.
3. Andrew J. Evans , Peter A. Humphrey, Jay Belani – Large cell neuroendocrine carcinoma of Prostate ; American Journal of Surgical Pathology ;Vol-30; June 2006; 684-693
4. Andrew R. Dodson, christopher s. Foster – The role of immunohistochemistry in the problematic prostate biopsy; Recent advances in histopathology -21, 105-136
5. Angelo M. Demarzo, Elizabeth A. Platz, Jonathan I. Epstein – A working group classification of focal prostate atrophy lesions. American Journal of Surgical Pathology ;Vol-30; October 2006; 1281-1291.
6. Anil V. Parwani, Cameron Marlow, Angelo M. Demarzo, - Immunohistochemical staining of Precursor forms of Prostate – specific antigen in metastatic prostate cancer; American Journal of Surgical Pathology ;Vol-30; October 2006; 1231-1236
7. Anil V. Parwani, Mehsati Herawi, Jonathan I. Epstein – Pleomorphic giant cell adenocarcinoma of the Prostate: Report of 6 cases; American Journal of Surgical Pathology ; October 2006; Vol – 30; 1254-1259

8. Ayala AG, VAE YR, Babain R, Troncoso P, Grignon DJ – The prostate ;American Journal of Surgical Pathology ; 13; 21-27, 1989
9. Badenoch A – Benign enlargement of the prostate ;Trans med sco; London 86: 1970; 34-40
10. Bostwick DG, Aruin MB, Dundone P, Marsh w, Scholtx DS- Architectural patterns of high grade prostatic intraepithelial neoplasia.; Human pathology;24; 1993; 298-310
11. Christopher D. M. Fletcher – Diagnostic histopathology of tumours; Second edition; vol – 1 & 2
12. David G. Bostwick, John N. Eble – Urologic surgical pathology; 343-422
13. David J. Dabbs MD – Diagnostic immunohistochemistry; second edition; 509-541
14. Donna E. Hansel and Jonathan I . Epstein – Sarcomatoid carcinoma of the Prostate: A Study of 42 cases. American Journal of Surgical Pathology ;Vol-30;October 2006; 1316-1321
15. Ecaterina F Tamas and Jonathan I. Epstein – Prognostic significance of paneth cell –like neuroendocrine differentiation in adenocarcinoma of the prostate American Journal of Surgical Pathology , Vol 30; August 2006; 980-985
16. Epstein J, Fynhur J – Acid mucin in the Prostae can it differentiate adenosis from adenocarcinoma?; Human Pathology; 23; 1992; 1321-1325
17. Ferdinandusse S, Denis S, Claytonn P T, et al.- Mutations in the genes encoding peroxisomal alpha – methylacyl – coa racemase cause adult onset sensory motor neuropathy, Nat Genet 2000; 24; 188 - 191

18. George J. Netto and Jonathan I. Epstein –Widespread High-grade Prostatic intraepithelial neoplasia on prosatic needle biopsy: A significant likelihood of subsequently diagnosed adenocarcinoma; American Journal of Surgical Pathology ;Vol-30;September 2006; 1184-1188
19. Gleason DF – Classification of prostatic carcinoma; Cancer chemotherapy ; 50; (1996) ;125-128
20. Goldstein N, Qian J, Bostwick DG - Mucin expression in atypical adenomatous hyperplasia of the prostate; Human pathology; 1995; 887-891
21. Green Lf, Farrow GM, RAUHS JM, TOMERA FM –Prostatic adenocarcinoma of ductal origin; Journal of urology 121; 1979; 303-305
22. Gregory M.Sheehan, Bhaskar U.S LKalllakury, Chirstine E – Loss of claudins 1 and 7 and expression of claudins 3 and 4 correlate with prognostic variables in prostatic adenocarcinomas, Human pathology, (38); (2007) ;564 – 569
23. Hukill P.B and Vinocle R.A – Histochemistry of mucus and other polysaccharides in tumour carcinoma of the prostate; Lab Ivest; 16; 1967; 395-406.
24. Humphrey PA- Mucin in severe dysplasia in the prostate; Surgical pathology 4; 1991; 137-143
25. Ivan Damjanov, James Linder – Anderson's pathology; Tenth edition; Vol 1 & 2
26. Jeffrey T.Schowinsky and Jonathan I. Epstein - Distorted rectal tissue on prostate needle biopsy: A mimicker of prostate cancer.

American Journal of Surgical Pathology, Vol 30; July 2006; 866-870.

27. Jiang z, Woda BA, Wucl, Yang X J – Discovery and clinical application of a novel prostate cancer marker- alphamethyl co A racemase; American Journal of surgical pathology; 2004;34;275-289
28. John D Bancroft, Marilyn Gamble – Theory and practice of histological techniques; fifth edition.
29. John N.Eble, Guido sauter, Jonathan I. Epstein & ISabell A. Sesterhenn – WHO Classification of tumours, Tumours of the urinary system and male genital organs (2004) 160 -214.
30. Jonathan I. Epstein, John srigley, David Grignon – Recommendations for the reporting of prostate carcinoma, Human pathology, (38); (2007); 1305- 1309
31. Jorge L. Yao, Ralph Madeb, Patricia Bourne, - Small cell carcinoma of the Prostate : An immunohistochemical study, American Journal of Surgical Pathology ;Vol-30; June 2006; 705-712
32. Juan Rosai ,Rosai and Ackerman's surgical pathology; Ninth edition; Vol 1 & 2
33. Kyllikki Haapala, Tuula kuukasjarvi, Eija Hyytinen – Androgen receptor amplification is associated with increased cell proliferation in prostate cancers, Human Pathology,(2007) 38; 474-478
34. Lars Holmberg, Anna Bill – Axelsen, Hans Garmo, - Prognostic markers under watchful waiting and radical prostatectomy,

Hematology/ Oncology clinics of North America; August 2006; Vol -20; 845-856

35. Latif A.Waja, Jodie palmer, Ladan fazli:- Comprehensive expression analysis of L-dopa decarboxylase and established neuroendocrine markers in neoadjuvantt hormone- treated versus varying Gleason grade prostate tumours;Human pathology, (38); (2007) ;161-170
36. Luna- Mores, Florez P. Ayala, A Diaz F, Santos – Neutral and acid mucins in carcinoma and AAH of the prostate pathology: Research& Practice:193(4), 1997, 291-298
37. Mc Mahon, M.J and Thomas G.H - Morphological changes of benign prostatic hyperplasia in culture,British Journal of cancer; 27; 323-335
38. Mehsati Herawi and Jonathan I. Epstein – Specialized stromal tumours of the prostate : A study of 50 cases; American Journal of Surgical Pathology ;Vol-30; June 2006; 694-704
39. Mehsati Herawi, Angelo M.Demarzo, Jonathan I.Epstein – Expression of CDX 2 in benign tissue and adenocarcinoma of the prostate, Human pathology (38); (2007); 72-78
40. Mehsatti Herawi, Elizabeth A. Montgomery and Jonathan I. Epstein – Gastrointestinal stromal tumours (GIST) on prostate needle biopsy: A clinicopathologic study of 8 cases. American Journal of Surgical Pathology ;Vol-30; November 2006; 1389-1395
41. Mellinger, G.T., Gleason D and Bailar, J III - The histology and prognosis of prostatic cancer; Journal of Urology; 97; (1967) ;331-337

42. Ming-Tse sung, Zhong Jiang, Rodolfo Montironi –Alpha methyl acyl co –A racemase (P504S)/34BE12/P63 triple cocktail stain in prostatic adenocarcinoma after hormonal therapy, Human Pathology (2007) 38; 332 – 341

43. Nobuyuki yanagisawa, Rile Li, David Rowkey – stromogenic prostatic carcinoma pattern in needle biopsy predicts biochemical recurrence – free survival in patients after radical prostatectomy,Human pathology (38) ;(2007) ;1611 – 1620

44. Oesterlicg JE – Benign prostatic hyperplasia. Medical and minimally invasive treatment options; J. England J Med; 332; 1995, 99-109

45. R. Montorini, D. Thompson, P H Bartets – Premalignant lesions of the prostate; Recent advances in histopathology-20; 131-146

46. Rile Li, sibel Erdamar, Hong Dai, Thomas M.Wheeler- Forkhead protein FKHR and its phosphorylated form P-FKHR in human prostate cancer, Human Pathology (2007) 38; 1501 – 1507

47. Ro, Grignon, Aruin, Ayala – Atlas of surgical pathology of the male reproductive tract.

48. Robbins and cotran pathologic basis of disease, 7th edition.

49. Robert H. Young, John R. Srigley, Mahul B. Arvin, Thomas M.Ulbright, Antonio L.Cubilla – Tumours of the prostate gland, seminal vesicles, male urethra and penis, Armed forces institute of pathology, Atlas of tumour pathology, Third series, Fascicle 28.

50. Satish Jain, Shubh Mahindru S. Jain – Prostate specific antigen (PSA), Journal of postgraduate medical education, training and research. March – April 2007, Vol 11, 12-16.

51. Sentinelli S - Mucins in prostatic intraepithelial neoplasia and prostatic carcinoma, Histochemistry 22 (3); March 1993; 271-274
52. Siamak Daneshmand, Marcus L. Quek, Edlin, Charlotte – Glucose – regulated protein GRP78 is up-regulated in prostate cancer and correlates with recurrence and survival, Human pathology (2007) 38; 1547 – 1552
53. Stacey E. Mills ,Sternberg's diagnostic surgical pathology; Fourth edition; Vol 1 & 2
54. Stanley S. Raphael – Lynch's Medical Laboratory technology – Third edition – 1976, Volume 1 & 2
55. Steven G. Silverberg, Principles and practice of surgical pathology and cytopathology; Fourth edition; vol 1& 2
56. Susan kasper and Michael S. Cookson – Mechanisms leading to the development of hormone resistant prostate cancer; Urologic clinics of North America; May 2006; Vol 23; 201-210
57. Svante R Orell, Gregory f stencett, Darrel whitaker –Fine needle aspiration cytology;. 4th edition; 366-373
58. Sven perner, Matthias D.Hofer, Robert kim, Rajal B.Shah, - Prostate specific membrane antigen expression as a predictor of prostate cancer progression, Human pathotogy,(38) ;(2007); 696 – 701
59. Walsh, Retik, Stamey, Vaughan – Campbell's urology, 6th edition, Volume 1 & 2
60. Zha S, ferdinandusse S, Hicks J L etal – Peroxisomal branched chain fatty acid beta – oxidation pathway is upregulated in prostatic cancer. Prostate, 2005; 63; 316-323